



August 02, 2013

LCDR Christopher Steele
Office of Naval Research
Code 34 – Warfighter Performance
875 N. Randolph St.
Arlington, VA 22203-1995

Subject: Final Report of the National Marrow Donor Program®

Reference: Grant #N00014-10-1-0204 between the Office of Naval Research and the National Marrow Donor Program

Dear LCDR Steele:

In accordance with the requirements of the Referenced Cooperative Agreement, the enclosed subject document is provided as the Final Report for each statement of work task item of the Grant for the period of March 01, 2010 through February 28, 2012.

With this submittal of the Final Report, the National Marrow Donor Program has satisfied the all reporting requirements of the above referenced Grant.

Should you have any questions as to the scientific content of the tasks and the performance activity of this progress report, you may contact our Chief Medical Officer – Dennis Confer, MD directly at 612-362-3425.

Please direct any questions pertaining to the Grant to my attention (612-362-3403 or at cabler@nmdp.org).

Sincerely,

Carla Abler-Erickson, M.A.
Contracts Manager

Enclosure: One (1) copy of subject document

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National Marrow Donor Program® N00014-10-1-0204

**Development of Medical Technology for
Contingency Response
To Marrow Toxic Agents**

FINAL REPORT

March 1, 2010 – March 31, 2012

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HLA Typing for Bone Marrow Transplantation
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ACRONYM LIST

AABB	American Association of Blood Banks
AAFA	African American (NMDP race code)
ABD	Antigen Binding Domain
AC	Apheresis Center
AFA	African American
AFB	African
AGNIS®	A Growable Network Information System
AIM	Ancestry Informative Markers
AINDI	South Asian
AISC	American Indian South or Central
ALANAM	Alaska Native or Aleut
ALDH	Aldehyde Dehydrogenase
ALDHbr	Aldehyde Dehydrogenase bright
ALL	Acute Lymphoblastic Leukemia
AMIND	North American Indian
AML	Acute Myelogenous Leukemia
API	Asian Pacific Islander
ARC GIS	ArcGIS is a brand name: GIS = Geographical Information System
ASBMT	American Society for Blood and Marrow Transplantation
ASH	American Society of Hematology
ASHG	American Society of Human Genetics
ASHI	American Society for Histocompatibility and Immunogenetics
ASPR	Assistant Secretary for Preparedness and Response
B2B	Business to Business
BARDA	Biomedical Advanced Research and Development Authority
B-LCLs	B-Lymphocytic Cell Lines
BMDW	Bone Marrow Donors Worldwide
BMT	Bone Marrow Transplant/Transplantation
BMT CTN	Blood and Marrow Transplant - Clinical Trials Network
BODI	Business Objects Data Integrator
BRIDG	Biomedical Research Integrated Domain Group
caDSR	Cancer Data Standards Repository
CARB	Black Caribbean
CARHIS	Caribbean Hispanic
CARIBI	Caribbean Indian
CAU	Caucasian
CB	Cord Blood
CBB	Cord Blood Bank
CBT	Cord Blood Transplantation
CBU	Cord Blood Unit
CC	Collection Center

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CEM	Certified Emergency Manager
CEO	Chief Executive Officer
CFO	Chief Financial Officer
CFU	Colony Forming Unit
CIBMTR®	Center for International Blood & Marrow Transplant Research
CIO	Chief Information Officer
CML	Chronic Myelogenous Leukemia
CMO	Chief Medical Officer
CRIS	Computerized Repository Inventory System
CSF	Colony Stimulating Factors
CT	Confirmatory Testing
CTA	Clinical Trial Application
CTAC	Clinical Trials Advisory Committee
DAIT	Division of Allergy, Immunology, and Transplantation
DC	Donor Center
DHHS	Department of Health and Human Services
DKMS	Deutsche Knochenmarkspenderdatei
DNA	Deoxyribonucleic Acid
DoD	Department of Defense
D/R	Donor/Recipient
DR	Disaster Recovery
DHHS	Department of Health and Human Services
DQ	Data Quality
DNA	Deoxyribonucleic Acid
DR	Disaster Recovery
D/R	Donor/Recipient
EBMT	European Group for Blood and Marrow Transplantation
EC	Ethics Committee
EFI	European Federation for Immunogenetics
ELISA	Enzyme-linked Immunosorbant Assay
EM	Expectation Maximization
EMDIS	European Marrow Donor Information System
ESRI	Environmental Systems Research Institute
FACS	Fluorescent Activated Cell Sorting
FILII	Filipino
FLOCK	Flow Cytometry Analysis Component
FY	Fiscal Year
GETS	Government Emergency Telecommunications Service
GCSF	Granulocyte-Colony Stimulating Factor (also known as filgrastim)
GIS	Geographic Information System
GM-CSF	Granulocyte Macrophage Colony Stimulating Factor
GVHD	Graft vs. Host Disease

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GWAS	Genome Wide Association Studies
Gy	Gray-measure of dose of irradiation
HAWI	Hawaiian or other Pacific Islander Unspecified
HC	Hematopoietic Cell
HCT	Hematopoietic Cell Transplantation
HHQ	Health History Questionnaire
HHS	Health and Human Services
HIS	Hispanic
HIV	Human Immunodeficiency Virus
HLA	Human Leukocyte Antigen
HML	Histoimmunogenetics Mark-up Language
HR	High Resolution
HRSA	Health Resources and Services Administration
HSC	Hematopoietic Stem Cell
HSCT	Hematopoietic Stem Cell Transplant
IBMTR	International Bone Marrow Transplant Registry
IBWC	Immunobiology Working Committee
IDM	Infectious Disease Markers
Ig	Immunoglobulin
IHIWS	International Histocompatibility Work Shop
IHWG	International Histocompatibility Working Group
IIDB	Immunobiology Integration Database
IIMMS	International Immunomics Society
IMGT	ImMunoGeneTics
ImmPort	Immunology Database and Analysis Portal
IPD	Immuno Polymorphism Database
IPR	Immunobiology Project Results
IRB	Institutional Review Board
IS	Information Services
IT	Information Technology
JAPI	Japanese
KIR	Killer Immunoglobulin-like Receptor
KIR-DS	Killer Immunoglobulin-like Receptor Donor Selection
KORI	Korean
LD	Linkage Disequilibrium
LTA	Lymphotoxin Alpha
MALDI-TOF	Matrix-Assisted Laser Desorption/Ionization – Time Of Flight
MCW	Medical College of Wisconsin
MD	Medical Doctor
MDACC	MD Anderson Cancer Center
MDS	Myelodysplastic Syndrome
MENAF	MidEast/North Coast of Africa

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MHC	Major Histocompatibility Complex
MICA	MHC Class I-Like Molecule, Chain A
MICB	MHC Class I-Like Molecule, Chain B
mHA _g	Minor Histocompatibility Antigen
MOU	Memorandum of Understanding
MRD	Minimal Residual Disease
MSKCC	Memorial Sloan-Kettering Cancer Center
MSWHIS	Mexican or Chicano
NAM	Native American
NAMER	North American
NCBI	National Center for Biotechnology Information
NCBM	National Conference of Black Mayors
NCHI	Chinese
NCI	National Cancer Institute
NECEP	New England Center for Emergency Preparedness
NHLBI	National Heart Lung and Blood Institute
NIAID	National Institute of Allergy and Infectious Diseases
NIH	National Institutes of Health
NIMA	Non-inherited maternal antigen
NK	Natural Killer
NL	Netherlands
NLM	National Library of Medicine
NMDP®	National Marrow Donor Program
NST	Non-myeloablative Allogeneic Stem Cell Transplantation
OCP	Operational Continuity Plan
ONR	Office of Naval Research
PA	Physician's Assistant
PBSC	Peripheral Blood Stem Cell
PCR	Polymerase Chain Reaction
PI	Principal Investigator
QC	Quality control
RadCCore	Radiation Countermeasures Center of Research Excellence
RCI	Resource for Clinical Investigations
RCI BMT	Resource for Clinical Investigations in Blood and Marrow Transplantation
RD Safe	Related Donor Safety
REAC/TS	Radiation Emergency Assistance Center/Training Site
REMM	Radiation Event Medical Management
RFP	Request for Proposal
RFQ	Request for Quotation
RITN	Radiation Injury Treatment Network
RT-PCR	Reverse Transcriptase-Polymerase Chain Reaction
SAA	Severe Aplastic Anemia

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SBT	Sequence Based Typing
SCAHIS	South/Central American Hispanic
SCAMB	Black South or Central America
SCSEAI	Southeast Asian
SCT	Stem Cell Transplantation
SCTOD	Stem Cell Therapeutics Outcome Database
SG	Sample Group
SNP	Single Nucleotide Polymorphism
SOP	Standard Operating Procedure
SRG	Survey Research Group
SSO	Sequence Specific Oligonucleotides
SSP	Sequence Specific Primers
SSOP	Sequence Specific Oligonucleotide Probes
STAR®	Search, Tracking and Registry
TBI	Total Body Irradiation
TC	Transplant Center
TNC	Total Nucleated Cell
UCB	Umbilical Cord Blood
UCBT	Umbilical Cord Blood Transplant
UI	User Interface
UML	Unified Modeling Language
URD	Unrelated Donor
US	United States
USB	Universal Serial Bus
VP	Vice President
VIET	Vietnamese
WebEOC®	Web-based Emergency Operations Center
WGA	Whole Genome Amplification
WHO	World Health Organization
WHO-REMPAN	World Health Organization, Radiation Emergency Medical Preparedness and Assistance Network
WMDA	World Marrow Donor Association
WU	Work-up
XML	Extensible Markup Language
ZKRD	Zentrales Knochenmarkspender – Register für die Bundesrepublik Deutschland

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Executive Summary

In 1986, Congress appropriated funds to begin development of the National Bone Marrow Donor Registry. Today, 26 years later, the National Marrow Donor Program (NMDP), as the contractor for the Registry, has built a racially diverse donor registry of 9.5 million donors, facilitated more than 50,000 hematopoietic stem cell transplants, developed comprehensive research programs to improve post-transplant outcomes, and established a network of transplant centers (TCs) capable of treating casualties resulting from military or terrorist actions, as well as patients suffering from leukemia, aplastic anemia, and other life-threatening diseases.

Contingency Preparedness Planning

During this funding period the Radiation Injury Treatment Network® (RITN) was expanded and further developed. Physician and staff education was made available through multiple venues from self-paced online training to instructor led courses and new partnerships were forged with federal agencies, as well as with private and non-profit organizations.

The NMDP's organizational resiliency was improved by conducting a drill that tested the Operational Continuity Plan and ensured staff would be able to work from a non-NMDP remote location and also by the acquisition of critical equipment.

Rapid Identification of Matched Donors

Published research data have clearly defined the relationship between Human Leukocyte Antigen (HLA) matching and optimal patient outcomes following unrelated adult donor transplantation. Continually working to increase the genetic diversity of the Registry helps to ensure that more patients will be able to locate a suitably matched stem cell product for a transplant. During NMDP's 2010 Fiscal Year (10/01/2009-09/30/2010), NMDP donor centers (including DoD) and recruitment groups recruited 270,284 minority race and 305,624 Caucasian donors, which were typed at minimum for HLA-A, B and DRB1. Navy funding contributed to the addition of 145,553 NMDP-recruited donors (excluding DoD). This added a culturally diverse group of new donors to the Registry.

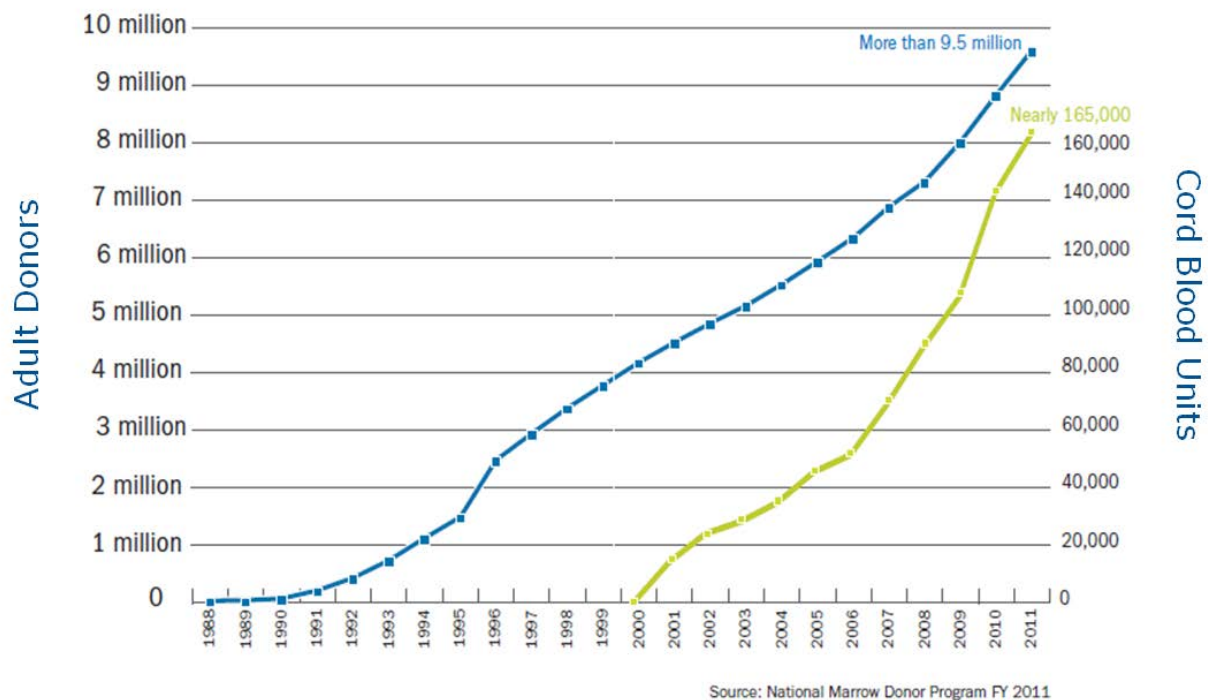
Continued advances in laboratory methods and supporting equipment have positively impacted the level of typing resolution for newly recruited volunteer donors. As of September, 2010:

- 96% of new donors received higher than intermediate HLA-A, B typing
- 45% of new donors received intermediate HLA-C typing
- 100% of new donors received higher than intermediate HLA- DRB1 typing

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- Blinded quality control testing accuracy rate was 99.88%, exceeding the project requirement of $\leq 2.0\%$.
- On-time testing completion rate was 98.8%, meeting the project requirement of a minimum of 90.0% of typing results reported within 14 days of shipment of samples.
- All new donors were typed at HLA-A, B, and DRB1. 34% of new donors were also typed at HLA-C. All typing was at intermediate resolution.
- The cost of HLA typing continues to decrease as technology improves; during the period March 2010 through August 2010 the average price per sample was approximately \$37.60, compared to \$134.75 in 1997, which represents a decrease of over 70%.

Be the Match Registry Growth: Adult Donors and Cord Blood Units



A study designed to evaluate the benefit of adding HLA-DRB1 to selected HLA-A, B only typed donors was completed during this award period. HLA-A, B only typed adult donors comprise 12% of the NMDP's Be The Match Registry® and are infrequently used for searching patients.

- 3,735 AB only donors on 210 preliminary patient searches, with stored DNA samples, were DRB1 typed and results compared to the corresponding patient

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- Only two donors matched their respective patients at DRB1
 - One was an ultimate 8/10 (5/6), with A and C allele mismatches
 - The other went to transplant for the patient, representing an HLA phenotype that hadn't previously existed on the registry

Based on the low rate of success, the strategy of testing HLA-A, B only donors may be best used in conjunction with selection of the best potential mismatched donor to avoid delaying transplant.

During this grant period, a program was optimized, validated, and run to “validate and push” HLA typing probe results, making them available to systems using HapLogic. Donors in the registry prior to February 2007 had already been pushed and have been used for searches; with this effort, approximately 1.1 million donors from 2007 through November 2008 have validated primary data used in searches.

NMDP released an enhanced version of the HapLogic algorithm that provided increased precision and clarity including:

- 3 locus matching was increased to 5 locus matching
- x of 6 is now x of 8 or x of 10 predictions
- 5 broad race groups were expanded to 5 broad and 18 detailed race groups
- Ensuring visibility of NMDP's best-matched donors and cord blood units (CBUs)
- More precision for mismatch searches
- Better alignment with clinical practice
- Realized performance improvements to the HapLogic algorithm with a median search run-time of 35 seconds

A set of tools and methods were developed to assess the optimal registry size for the U.S. population. The tools utilize HLA haplotype frequency data to determine matching rates for the registry (adult donor and cord blood) under a variety of growth scenarios. The results were incorporated into several presentations and the first draft of a match-rate manuscript targeted to transplant physicians was drafted.

A new version of the “haplostats” tool (<http://haplostats.org>), which is used by laboratories and investigators worldwide to look up HLA types and present the corresponding haplotype frequency data in a way that can inform research and medical decision making, was released. As an enhancement to this software, we have identified a goal to generate automated maps of HLA haplotype frequencies projected on to maps of the US and the world. We have been working with software tools from Environmental Systems Research, Inc. (ESRI) to automate map production and have completed a prototype of desktop-scale map automation in an easy-to-read world view map.

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Immunogenetic Research

The high resolution HLA typing of paired donor and recipient samples continued to provide substantive data to increase the understanding of the impact of HLA matching on patient outcome. The project data were also used to assess genetic diversity within the NMDP transplant population and Registry, and fed into the HapLogic matching algorithm. Testing was completed on an additional 146 donor/recipient and 192 cord/recipient pairs during the project period, bringing the total enrolled to over 15,000. Typing at the HLA-DPB1 locus was added back to facilitate future studies of HLA-DPB1 matching. Presence/absence Killer Immunoglobulin-like Receptor (KIR) genotyping on 2DL1-5, 2DS1-5, 3DL1-3 and 3DS1 has continued. To date over 2100 pairs and 1180 additional donors have been typed for presence/absence of 14 KIR loci (2DL1-5, 2DS1-5, 3DL1-3 and 3DS1).

In order to continuously upgrade the Donor/Recipient Pairs Project, and include as many CBU/recipient pairs as possible, we have begun to include pairs with Whole Genome Amplified (WGA) DNA from the Repository. Samples of recipients and CBUs with limited blood or DNA samples in the research repository were selected and sent for WGA. WGA of these samples allowed for the inclusion of 98 CBU/recipient pairs in Sample Group (SG) 27.

The high-resolution HLA data generated through the project are routinely incorporated into all outcomes analyses performed by the Center for International Blood and Marrow Transplant Research (CIBMTR) to provide the best HLA typing and matching information possible. The project has developed the largest, fully validated pool of unrelated stem cell transplant donor-recipient HLA data in the world and is an unparalleled resource for transplant research. The data generated through the project have had a major impact on the evolution of the NMDP HLA matching requirements.

Current HLA matching guidelines for unrelated Hematopoietic Cell Transplantation (HCT) recommend avoidance of mismatches only within the antigen recognition site, i.e. exons 2 and 3 for HLA class I and exon 2 for HLA class II. This recommendation is based on the hypothesis that amino acid differences outside the antigen recognition site are not immunogenic. The Antigen Recognition Site Allo-reactivity Assessment Project will give insight into the allowable tolerance of matching needed outside of this binding region. Work was initiated to identify specific mismatches for evaluation in the project.

Clinical Research in Transplantation

Improving strategies to avoid and manage graft-versus-host disease (GVHD) is an essential step in improving the outcomes of transplantation and, consequently, the ability to incorporate transplantation as an effective therapy into a variety of settings, including contingency situations.

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The goal of the research activities funded through this grant has been to increase the understanding of the immunologic factors important in HCT.

During this grant period, the Resource for Clinical Investigations in Blood and Marrow Transplantation (RCI BMT) continued to work towards its goal to provide an avenue for investigators to obtain statistical and data management support for prospective trials and projects in HCT. The following key elements were completed:

- Clinical Trials Advisory Committee (CTAC) held two annual in-person meetings during this grant period. Both meetings occurred during the Tandem meetings, in February 2011 and 2012. This committee has been charged with providing scientific review and recommendations on clinical trial proposals. The committee reviewed two proposals and one was approved to move forward to protocol development. The CTAC recommended the second proposal not proceed because of concerns that accrual could not be achieved in a reasonable time period.
- Accrual was completed on the Adult Double Cord protocol for patients with hematologic malignancies on September 13, 2011. A total of twelve sites enrolled 56 patients during the course of this trial of which the last 20 occurred during this grant period. Staff continued working with sites to ensure all data was submitted, data queries were addressed and performed site monitoring.
- Staff continued to provide support to the Blood and Marrow Transplant Clinical Trials Network (BMT CTN) PBSC vs. Marrow Phase III trial. This support included managing the donor component of the study. Staff performed monitoring activities at the donor centers. During this grant period a total of 20 monitoring visits occurred with 320 of the 551 donor data sets were monitored.
- The Long-Term Donor Follow up (LTDFU) study opened October 1, 2010. During this grant the logistics and processes were established. Accrual to this study included donors who previously donated in addition to donors who are donating currently. As of the end of this grant period a total of 10,838 donors had consented to participate. This is 34% of the goal of 32,128 donors. The Survey Research Group (SRG) staff is responsible for the follow up contacts for the NMDP operated donor centers. During this grant period SRG completed 3,428 follow up calls.

Support of the Observational Research program included statistical hours for managing studies within the Immunobiology, GVHD, and Graft Sources Working Committees. During this grant period, staff performed proposal review, protocol development, data preparation, data analysis, and manuscript preparations. Details regarding the Immunobiology activities can be found in IID1.3 below. The GVHD and Graft Sources Working Committees published 11 manuscripts. During the grant period, staff supported progress on over 20 other studies.

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The Cord Blood Research subcommittee continued work on several ongoing projects. Work continued to validate the testing methodologies for the “Cord Blood Biomarkers for Engraftment” study at three centralized laboratories to ensure consistent inter-laboratory results. An observational study of single versus double cord blood transplants (CBT) in adults proceeded in collaboration with the CIBMTR Graft Sources Working Committee. An analysis of the impact of matching for non-inherited maternal antigens (NIMA) in mismatch CBT was implemented and completed. The results suggest that matching for NIMA may improve survival in HLA mismatched CBT for acute leukemia and were submitted for publication. A white paper detailing recommendations and guidelines for the assessment of new assays (potency or other assays) relevant to cord blood banking and cord blood evaluation for transplantation was completed and accepted for publication in Cytotherapy.¹ The laboratory staff from the cord blood banks worked with Stem Cell Technologies to assess the efficacy of STEMvision, an automated colony forming unit (CFU) enumeration instrument. Work began on the pilot study to assess a system for centralized flow cytometry based CD34 analysis in an effort to improve intra-laboratory variability observed through the NMDP proficiency testing program.

To further stimulate completion of immunobiology studies within the CIBMTR, grant funds were used to provide support to investigators that required supplemental funding to cover research sample access costs. One grant was awarded during the grant period. Grant funds also supported Immunobiology Working Committee (IBWC) leadership outreach activities to promote the activities and resources of the committee to the scientific community. The committee maintained a strong performance record with 10 abstracts, 9 publications (submitted or accepted) and collaboration on 3 grants completed in calendar year 2011. In addition, 6 new proposals were accepted by the IBWC during the BMT Tandem meetings in February 2012.

END – EXECUTIVE SUMMARY

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II.A. Contingency Preparedness – Hypothesis 1:

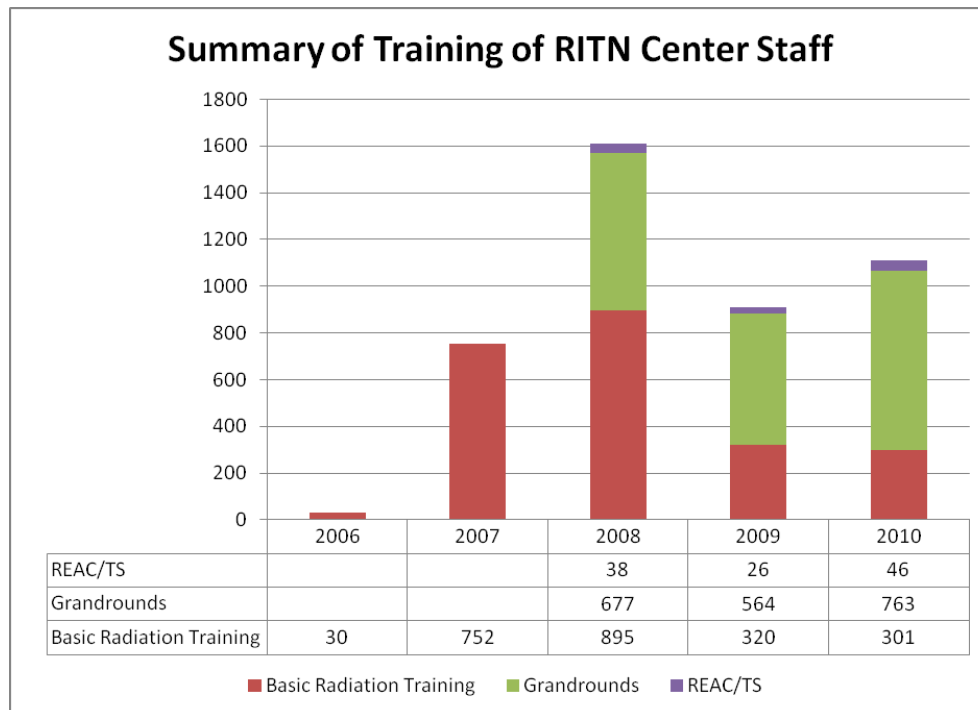
Recovery of casualties with significant myelosuppression following radiation or chemical exposure will be optimal when care plans are designed and implemented by transplant physicians

Aim A.1.1: Secure Interest of Transplant Physicians

In working to accomplish this Aim, the NMDP focused on the education of transplant physicians and their staff from Network centers affiliated with the Radiation Injury Treatment Network® (RITN).

These educational efforts have grown from a self-paced Basic Radiation Training course in 2006 to a standardized Acute Radiation Syndrome Medical Grand rounds curriculum and RITN center staff attendance at the Radiation Emergency Assistance Center and Training Site (REAC/TS) for instructor led training. During this performance period, over 1,000 staff received RITN training; a summary of training progress year to year since 2006 is displayed in Figure 1. The variance in the number of individuals receiving Basic Radiation Training between 2007 & 2008 versus subsequent years is due to the restriction of tracking only previously untrained staff; therefore the pool of untrained staff at RITN centers diminishes over time.

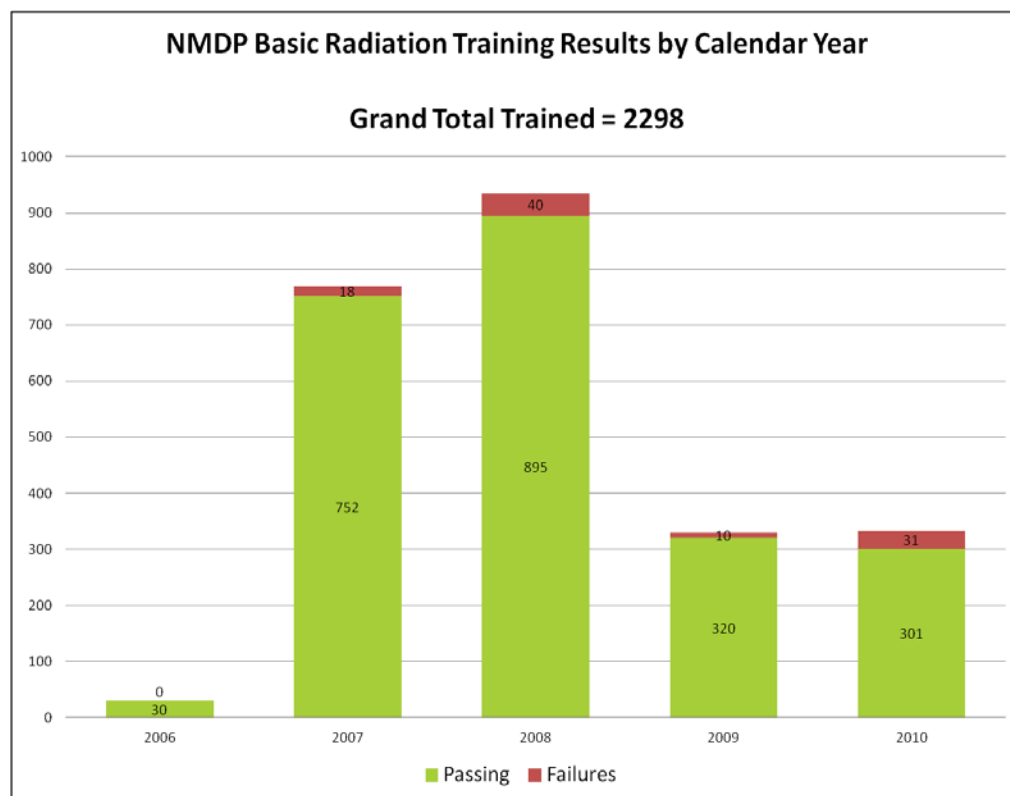
Figure 1: RITN center staff completing radiation treatment training from 2006-2010.



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RITN centers are required to complete annual training exercises to be considered in good standing and this has driven the need for multiple training options to ensure that the new, larger audiences can be brought into the fold, as well as providing variety for staff that participate each year. During this period of performance, over 300 RITN center staff successfully completed the Basic Radiation Training; overall 2,298 staff have successfully completed this training since 2006 (Figure 2).

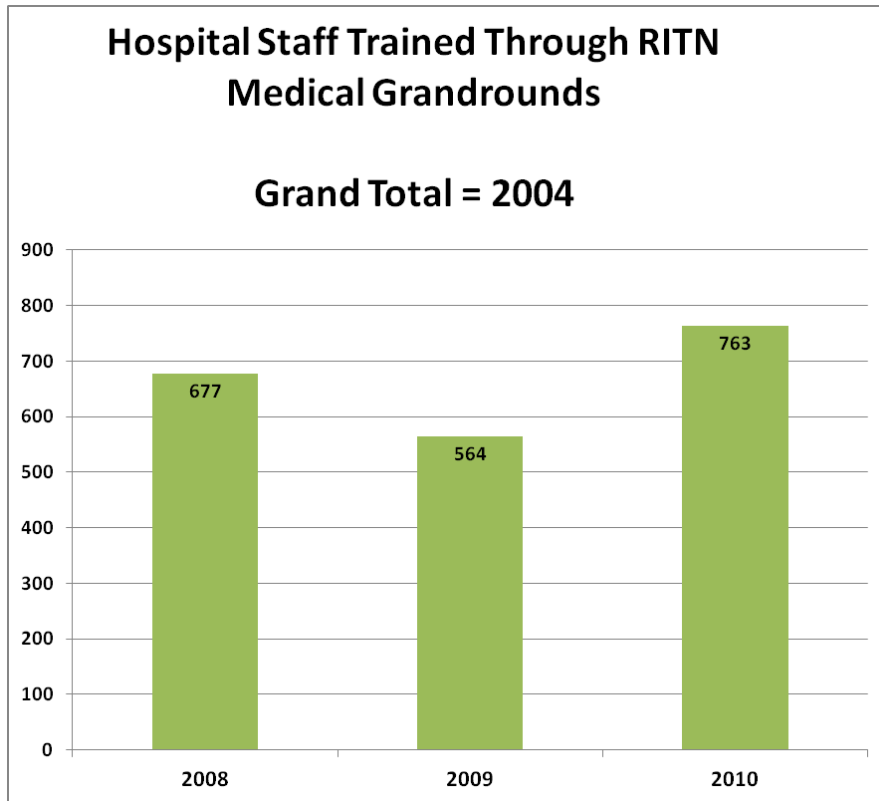
Figure 2: RITN center staff completing the Basic Radiation Training course during the period of performance.



During the Medical Grand rounds training on the treatment of Acute Radiation Syndrome, 763 RITN center staff were successfully trained; a total of 2,004 staff trained since 2008 when this course was implemented (Figure 3).

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Figure 3: RITN center staff receiving Medical Grand rounds training on the treatment of Acute Radiation Syndrome during the period of performance.



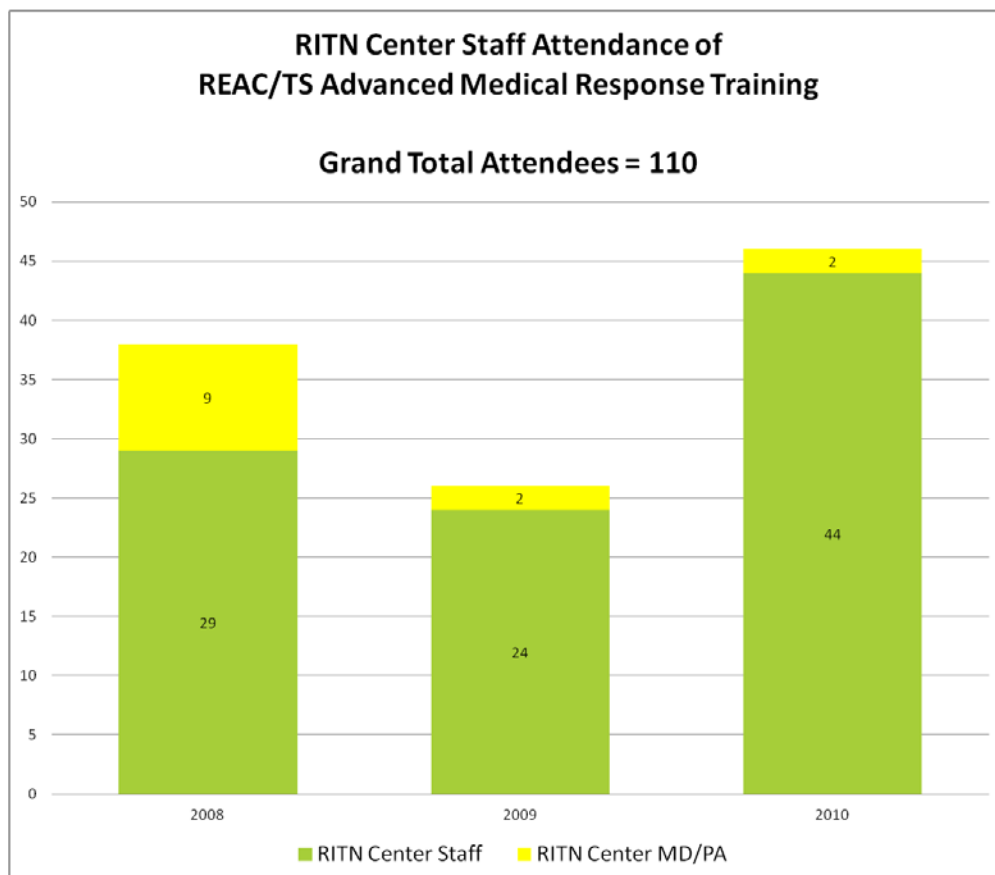
During this period of performance, two groups of RITN center staff totaling 46 people, (class size is limited to 25 maximum), traveled to Oak Ridge, Tennessee to attend the two day Advanced Medical Training on Radiation Emergency Medicine course provided at the REAC/TS facility (March 29-30, 2010 and September 9-10, 2010). The course covers a comprehensive set of topics including:

- Basic Health Physics & Radiation Protection: Part I
- A History of Serious Radiological Incidents: The Real Risk
- Health Physics & Contamination Control: Part II
- Radiation Detection, Monitoring & Protection Laboratory Exercise & Quiz
- Diagnosis & Management of Acute Radiation Syndrome
- Diagnosis & Management of Internal Contamination
- Diagnosis & Management of Acute Local Radiation Injury & Case Review: Yanango Peru
- Radiation Sources & Radiological Terrorism
- Radiation Emergency Area Protocol Demonstration
- Radiation Emergency Medical Management Drill
- Radiation Dose Estimations – Problem Solving Session

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Attendees earn 14 AMA PR Award Category 1 Continuing Medical Education Credits for attending the course. However, due to the location and course hours attendees typically must arrive the day prior to the course and depart the day following the completion, this increasingly has made the involvement of physicians difficult, as taking up to four days out of their schedule for training is a significant commitment. The RITN Executive Committee reviewed options to entice more physicians to attend this training in subsequent years. Since RITN began sending staff to this instructor led REAC/TS course, 110 staff have received this training (Figure 4).

Figure 4: RITN center staff completing the REAC/TS course during the period of performance.



Aim A.1.2: GCSF in Radiation Exposure

No funding was requested under this Aim for the 0204 budget cycle.

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Aim A.1.3: Patient Assessment Guidelines and System Enhancements

A tool was implemented to provide the ability to electronically contact the donors via email and allow them to update their contact information and complete an online Health History Questionnaire (HHQ) from the Do It Yourself Donor online platform. Information provided by the donor is securely transferred to the donor's record in the tool used to manage Donor Activity; facilitating reporting, storage and review of this information in established donor management systems.

Project Outcomes:

1. New versions of the tools used to manage Donor Activity have been implemented and continue to show favorable results with strong user feedback:
 - **Donors continue to be responsive to the online tools.** New Online HHQ functionality resulted in:
 - 8,471 **“Completed”** HHQs
 - 470 **“in process”** HHQs (between 10/1/09 – 09/30/10)
 - A 50% reduction in processing time per online HHQ has resulted in an overall time savings of 1624 hrs.
2. An Event Portal Workflow Management Application to manage contingency events was implemented and is in production for all Domestic NMDP Network Donor Centers, excluding the DoD, DKMS Americas, Gift of Life Registry, and Caitlyn Raymond Registry.

Key features:

- Ability to track preliminary event donors in a central screen, for purposes of donor management.
- Ability to import the preliminary event donors, as identified through the preliminary event daily report.
- Ability to export the preliminary event donors for purpose of supporting address validations, manual mail merges, or automated letter merges.

Key statistics gathered between 10/1/09 – 09/30/10 for the Event Portal:

- Completed 5,197 Preliminary Search HHQs
- Activated 2,141 Preliminary Search donors
- Average close date of 7 days on an Preliminary Search HHQ

Aim A.1.4: National Data Collection Model

No funding was requested under this Aim for the 0204 budget cycle.

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II.A. Contingency Preparedness – Hypothesis 2:

Coordination of the care of casualties who will require hematopoietic support will be essential in a contingency situation.

Aim A.2.1: Contingency Response Network

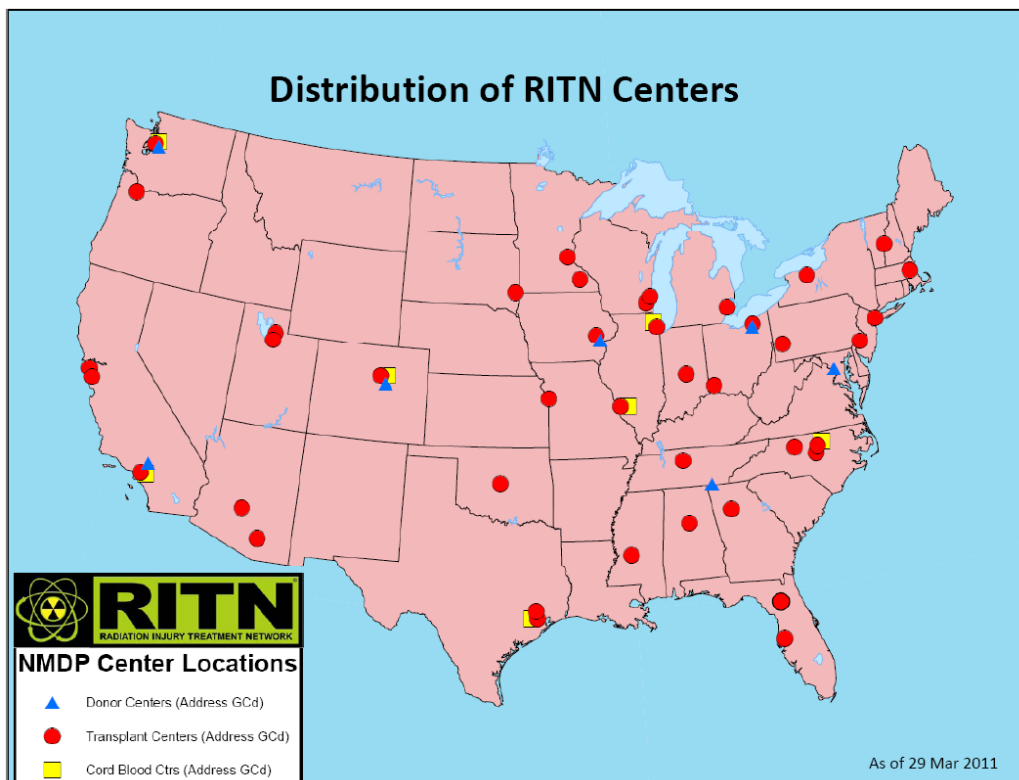
The RITN and its efforts were the focus of this Aim. The RITN was organized to provide comprehensive evaluation and treatment for victims of radiation exposure or other marrow toxic injuries. The RITN develops treatment guidelines, educates health care professionals, works to expand network membership, and coordinates situation response. The RITN is a cooperative effort of the NMDP and The American Society for Blood and Marrow Transplantation (ASBMT).

During this period of performance, the RITN consisted of 55 centers including:

- 41 transplant centers
- 7 donor centers
- 7 cord blood banks

The locations of the various RITN centers are shown in Figure 5.

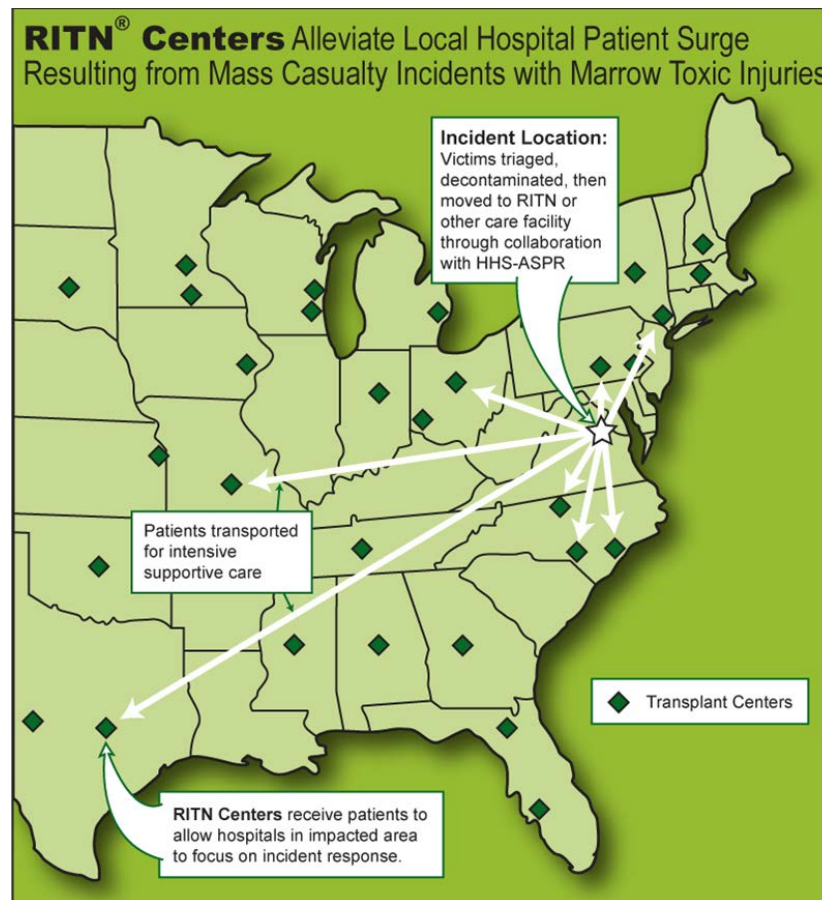
Figure 5: RITN Center Locations in the United States



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RITN centers are prepared to receive casualties from a mass casualty incident with marrow toxic injuries. However, RITN centers are not first responders. All participating centers voluntarily prepare to respond to an incident that occurs regionally, nationally, or internationally. The NMDP anticipates that the RITN centers will receive patients from other parts of the country to alleviate the medical load at the site of the incident and to provide the best marrow toxic injury care possible (see Figure 6 for overview of casualty flow).

Figure 6: RITN Center Casualty Flow Overview.

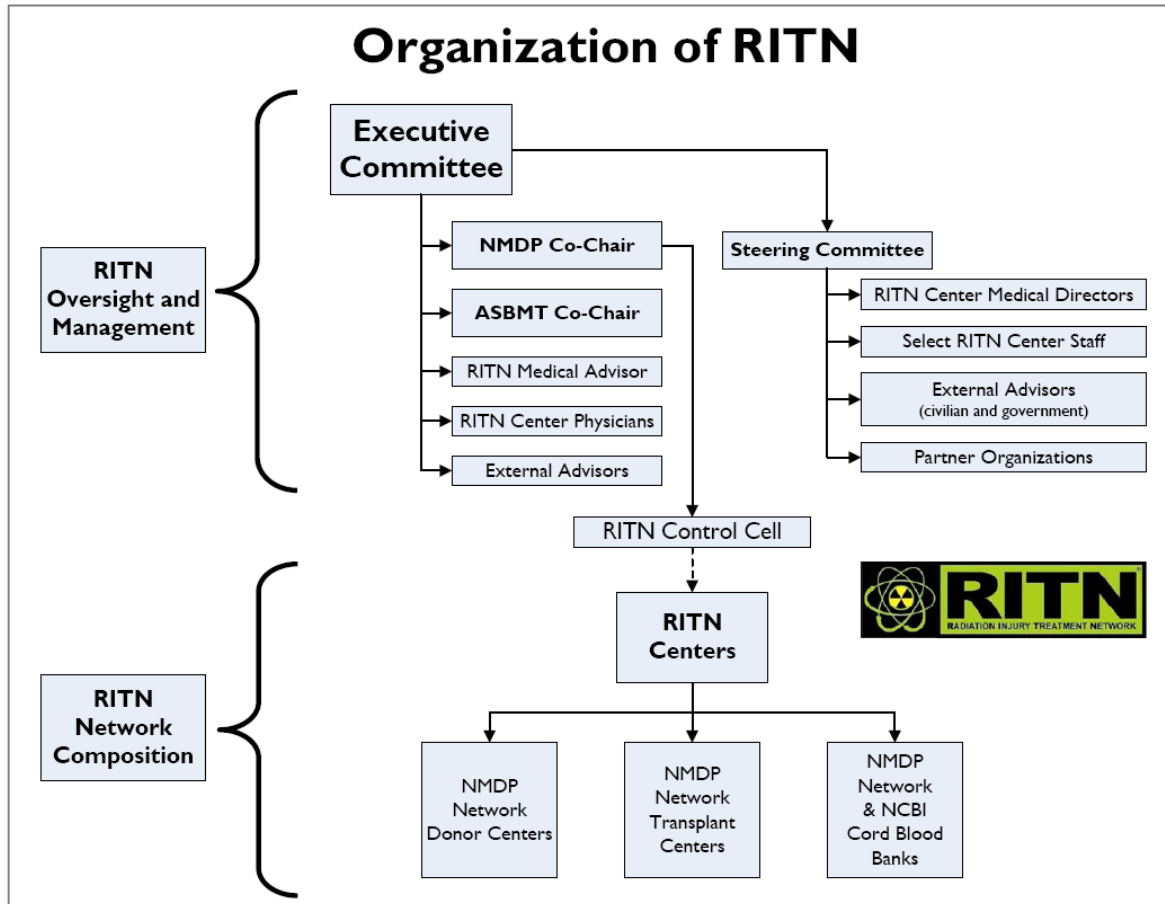


RITN Management

The daily operations of the RITN are managed by the RITN Control Cell (Figure 7), with oversight provided through an Executive Committee and a Steering Committee. The Executive Committee meets every other month via conference call to review current projects and plan for future activity. The Steering Committee meets annually at the BMT Tandem Meetings, bi-annually at the RITN Educational Conference and is composed of the medical directors of each RITN center (including Transplant Centers, Donor Centers and Cord Blood Banks) and partner organization representatives.

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Figure 7. Organization of RITN



During the period of performance the **Steering Committee** held three in person meetings:

- February 2011 - ASBMT/CIBMTR Tandem Meetings (Honolulu, HI)
- October 2011 – 2011 RITN Educational Conference (Chicago, IL)
- February 2012 - ASBMT/CIBMTR Tandem Meetings (San Diego, CA)

Outcomes of these meetings included:

- RITN Year in Review summarizing accomplishments and planned activity
- Overview presentations from industry about research and developments related to treatment for an prevention of marrow suppression

The **RITN Executive Committee** is chaired by the Chief Medical Officer of the NMDP and a representative from the ASBMT, and assisted by a Medical Advisor, other physicians and technical advisors:

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- Committee Chairs:
 - Co-Chair: Dennis Confer, MD
 - Co-Chair: Nelson Chao, MD
- RITN Medical Advisor:
 - David Weinstock, MD
- Committee Members:
 - Transplant Physician: Daniel Weisdorf, MD
 - Transplant Physician: John Chute, MD
 - ASBMT Representative: Julie Wilhauk, ARNP, AOCNP
 - ASBMT Alternate Representative: Robert Krawisz, MBA
 - RITN Program Manager: Cullen Case Jr., CEM

Executive Committee accomplishments during this period of performance include:

- Identification of possible bone marrow transplant programs to be invited to join RITN in the future
- Development of the agenda, content, and speakers for the 2011 RITN Educational Conference “State of the Science Workshop: Radiation Exposure, Medical Countermeasures and Treatment”
- Development of agendas for Steering Committee meetings and coordination of presentations by external subject matter experts at the Steering Committee meetings

A key partnership was established with the New England Center for Emergency Preparedness (NECEP) through a Memorandum of Understanding. As a result of this relationship, RITN was able to transfer from using Web-based Emergency Operations Center (WebEOC) as a Crisis Management Software program with annual maintenance costs to the Health Care Standard which is maintained and funded by NECEP.

Every month all RITN centers and partners are invited to a conference call where updates are provided on current projects as well as the “Rad in the News” is reviewed. “Rad in the News” is a summary of highlights with links to open source media reports about radiological related current events. In addition to these monthly meetings, two webinar presentations were offered for RITN centers and partners. The first webinar was a Best Practices review of how Memorial Sloan Kettering Cancer Center (MSKCC) implemented RITN at their hospital. The session included a review of the hurdles surmounted and key lessons learned during the MSKCC implementation. The second webinar was a RITN Year in Review. The webinar reviewed the annual highlights from new RITN centers, center participation in marrow toxic injury exercises, new partnerships established, and the Annual Tasks planned for the next year.

RITN Annual Tasks

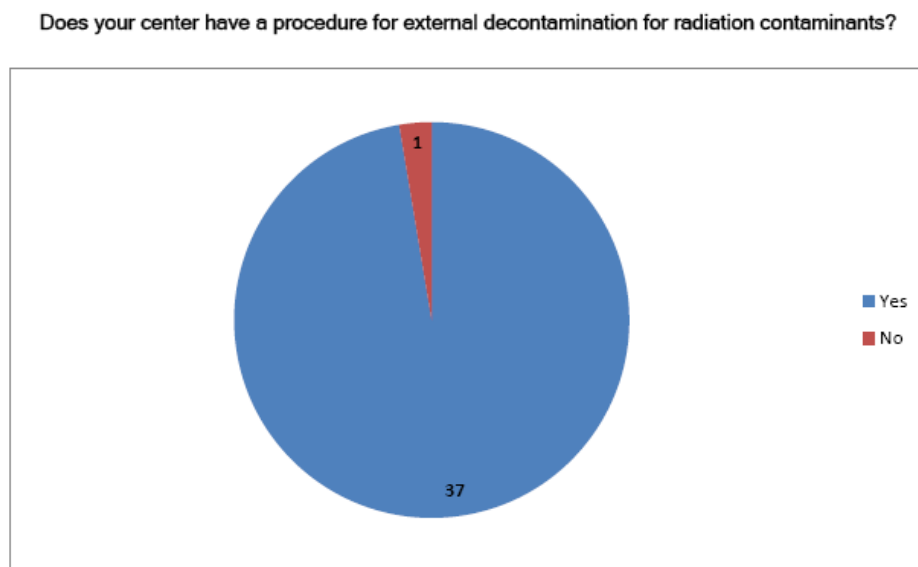
RITN centers are asked each year to complete a set of tasks in exchange for a small grant. Centers were asked to update their standard operating procedures, conduct a tabletop exercise, and conduct training of staff.

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The scenario for the tabletop exercise presented the detonation of a 10 kT groundburst improvised nuclear device in a major metropolitan area. Centers gathered a team of subject matter experts to review how they would respond to such a scenario; on average, transplant centers had 16 people participate in their exercise, donor centers had 17 people involved, and cord blood banks had 10 people participate in the exercise. Centers were stressed by the simulated arrival of 500 patients to their hospital for triage and care. It was not expected that all centers would be able to handle this large of a surge, but rather the exercise was designed to force them to review how they would handle such a surge. Centers were required to consider what organizations they would have to work with to distribute casualties for care and what new processes may be necessary for them to handle a potential disaster.

After reviewing the scenario, RITN centers answered a set of questions that were submitted for review. Some of the highlights from these responses to the tabletop exercise questions are shown in Figures 7-10.

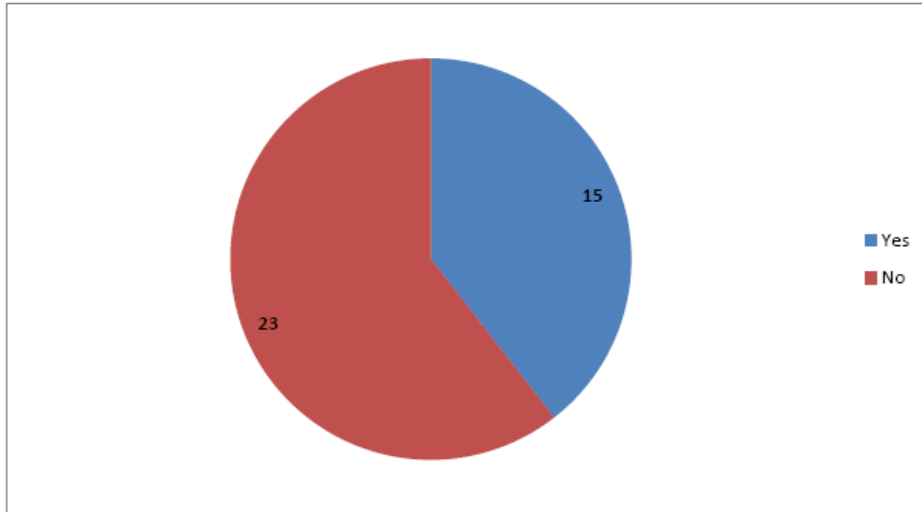
Figure 7. Selected responses from RITN Table Exercise



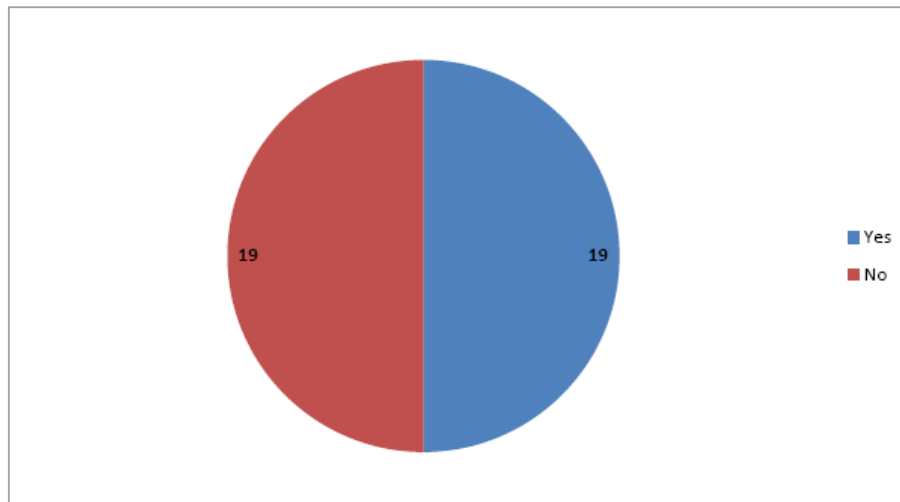
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Figures 8 & 9: Selected responses from RITN Table Exercise (continued)

Does your organization have radiation survey portals?

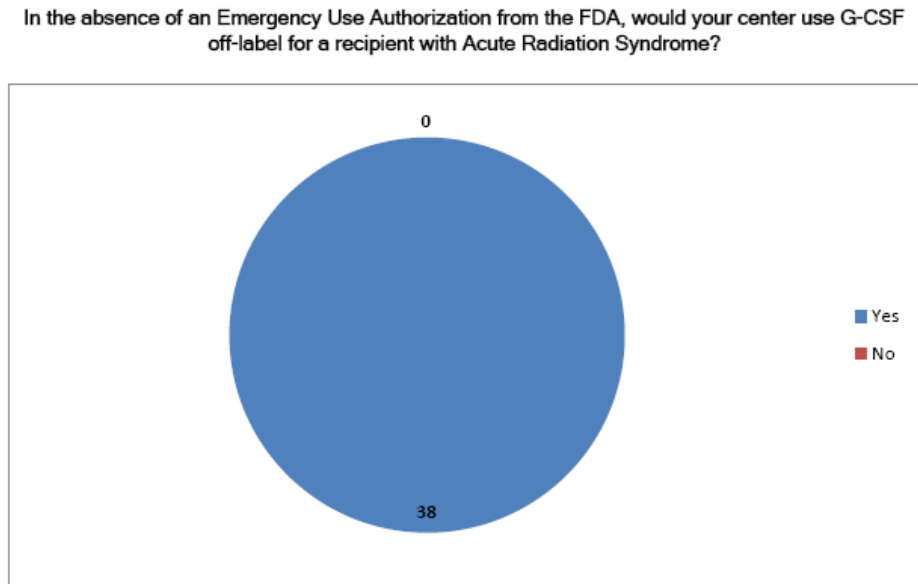


Does your organization have access through a partner organization to borrow radiation survey portals?



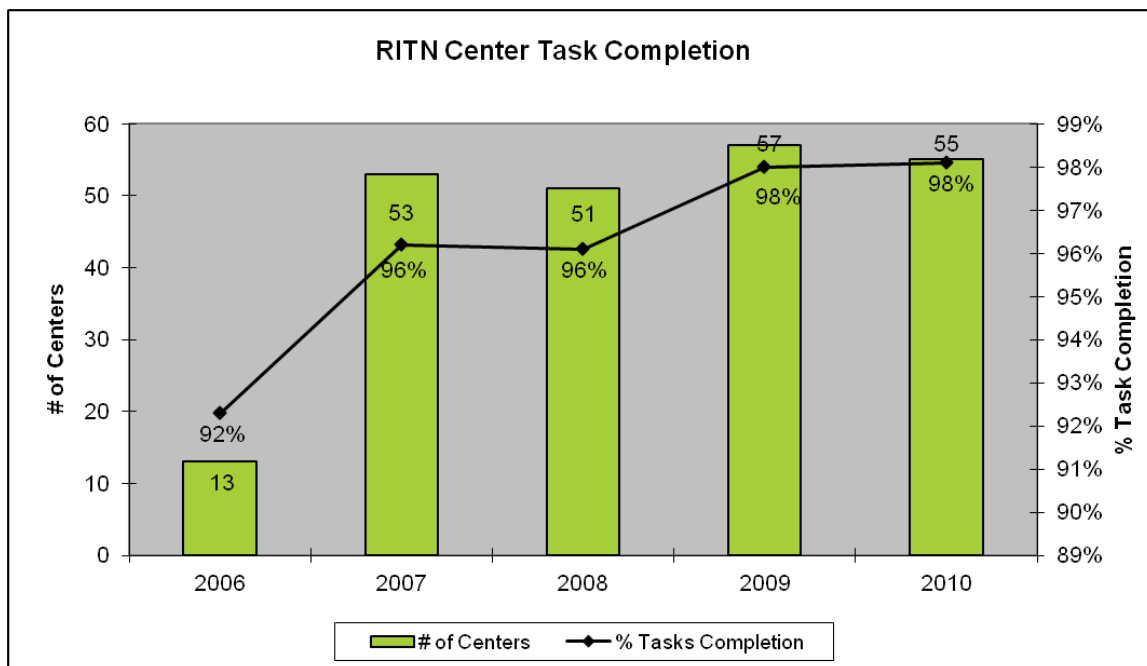
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Figure 10. Selected responses from RITN Table Exercise (continued)



During this grant period, 98% of the RITN centers completed all of their tasks, a slight improvement over the previous grant period (Figure 11).

Figure 11: RITN Center task completion by year.



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The RITN would not be able to successfully respond to a mass casualty incident without the support of partner organizations. The NMDP has carefully worked to develop and maintain key partnership relationships so that when an event occurs we are prepared to interface with the key response organizations.

RITN has developed two types of relationships with partner organizations; formal relationships that are documented through a Memorandum of Understanding (MOU) and informal relationships that are maintained through periodic collaboration.

RITN has established formal relationships through an MOU with:

- Office of the Assistant Secretary for Preparedness and Response, Department of Health and Human Services (ASPR-DHHS)
- ASBMT
- American Association of Blood Banks (AABB), through the AABB Inter-organizational Task Force for Disasters and Acts of Terrorism
- NECEP

RITN has developed informal partnerships with the following organizations:

- Radiation Countermeasures Centers of Research Excellence (RadCCORE) at Duke University
- The National Institutes of Health, The National Institute of Allergy and Infectious Diseases, Division of Allergy, Immunology and Transplantation (NIH-NAIAD-DAIT)
- Radiation Emergency Medical Management web portal (NIH-NLM-REMM)
- National Cancer Institute (NCI)
- Biomedical Advanced Research and Development Authority (BARDA)
- European Group for Blood and Marrow Transplantation (EBMT) - Nuclear Accident Committee
- The Radiation Emergency Medical Preparedness and Assistance Network of the World Health Organization (WHO-REMPAN)
- Radiation Emergency Assistance Center and Training Site (REAC/TS)
- American Hospital Association (AHA)
- American Medical Association (AMA)

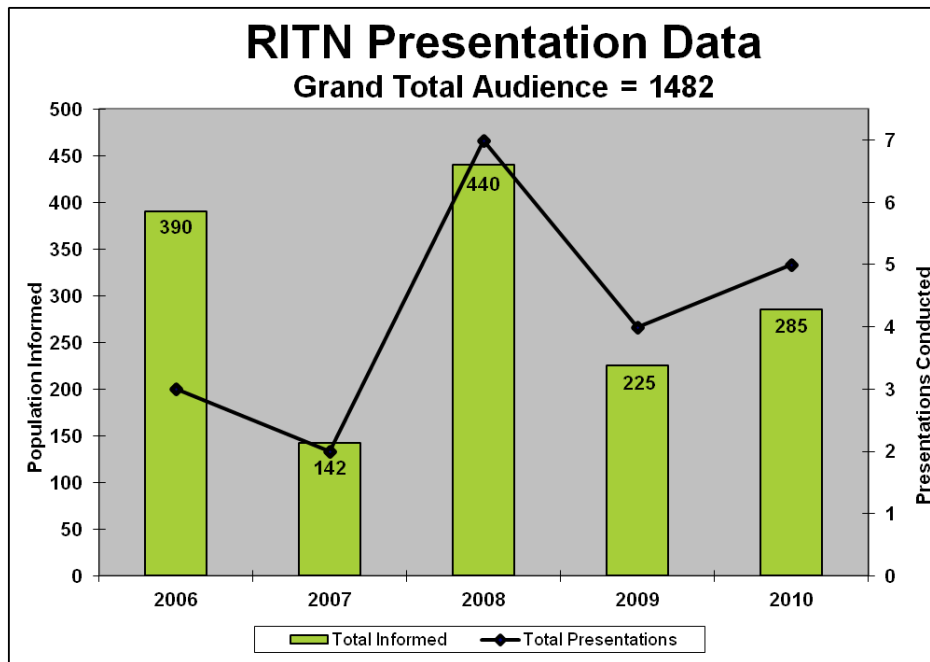
To increase the visibility of RITN and make new connections with additional organizations and agencies, overview presentations were given to various professional groups and government agencies (Figure 12). RITN overview presentations were delivered to the following groups during this period of performance:

- Emergency Management Summit in Washington, DC (presenter: Richard Hatchett, M.D.)
- New York City Department of Health Hospital Emergency Preparedness Program workshop in New York City, NY (presenter: Larry Dauer, PhD)
- Canadian National Mass Casualty Capacity Review in Ottawa, Canada (presenter: Cullen Case)
- AABB Disasters Task Force in Bethesda, MD (presenter: Cullen Case)

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- Iowa National Disaster Medical System Tabletop Exercise in Cedar City, IA (presenter: Colleen Chapleau)

Figure 12: RITN overview presentations from 2006-2010 noting the number of total attendees and presentations conducted.



Another avenue for increasing the visibility of RITN is through publications in peer reviewed journals. The RITN Executive Committee participated in three publications during this period of performance:

- Joel Ross, Cullen Case, Nelson Chao, et al. Radiation Injury Treatment Network (RITN): Healthcare professionals preparing for a mass casualty radiological or nuclear incident. *Int. J. Radiat. Biol.*, Vol. 87, No. 5, PrePublished Feb 2011
- Davids MS, Case C Jr, et al. Assessing Surge Capacity for Radiation Victims with Marrow Toxicity. *Biol Blood Marrow Transplant*, Oct 2010, 16(10):1436-41
- Davids MS, Case C Jr, et al. Medical Management of Radiation Victims in the United States, *Health Physics*, June 2010, 98 (6): 833-837

RITN preparedness and response activities during the period of performance included situational awareness emails, participation in a national exercise, development of a site assessment program, and contribution to a strategic plan presented to White House staff.

For approximately 90 days following the disaster at the Japanese Nuclear Power Plant in Fukushima, daily summary situational awareness reports were sent to RITN centers and RITN partner organizations. RITN received requests from the AHA to the Centers for Disease Control

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to receive copies of these reports, increasing the number of RITN partners and adding to RITNs visibility in the preparedness community.

36 RITN centers participated in the 2010 United States Homeland Security National Level Exercise which simulated the response to the detonation of an improvised nuclear device. RITN centers submitted capabilities reports that were rolled up and forwarded to the DHHS-Assistant Secretary for Preparedness and Response. The reports helped to increase awareness of RITN capabilities during the exercise.

To meet the educational needs of the growing network, a web based learning management system was procured that will allow RITN center staff to complete training online, including testing and receipt of their certificate of training completion. This system will also facilitate the delivery of just-in-time training that can be used by RITN centers, referral hospitals, or other partners that find a need for radiological medical treatment training.

To leverage knowledge gained from RITN center site visits and review of their standard operating procedures, RITN staff developed checklists for conducting site assessments of RITN centers to spread best practices across the network. These checklists reviewed five functional areas at each RITN center including:

1. Victim processing
2. Outpatient treatment of victims
3. Inpatient treatment of victims
4. Coordination with region, state, or federal agencies
5. Documentation Review

Assessments using the developed checklists did not begin until the following period of performance.

At the request of National Security Council (NSC) staff, the RITN Executive Committee developed a two year strategic plan that would allow RITN to increase its capacity to meet the needs of the Nation following a nuclear incident. This strategic plan was provided to NSC staff who briefed White House staff.

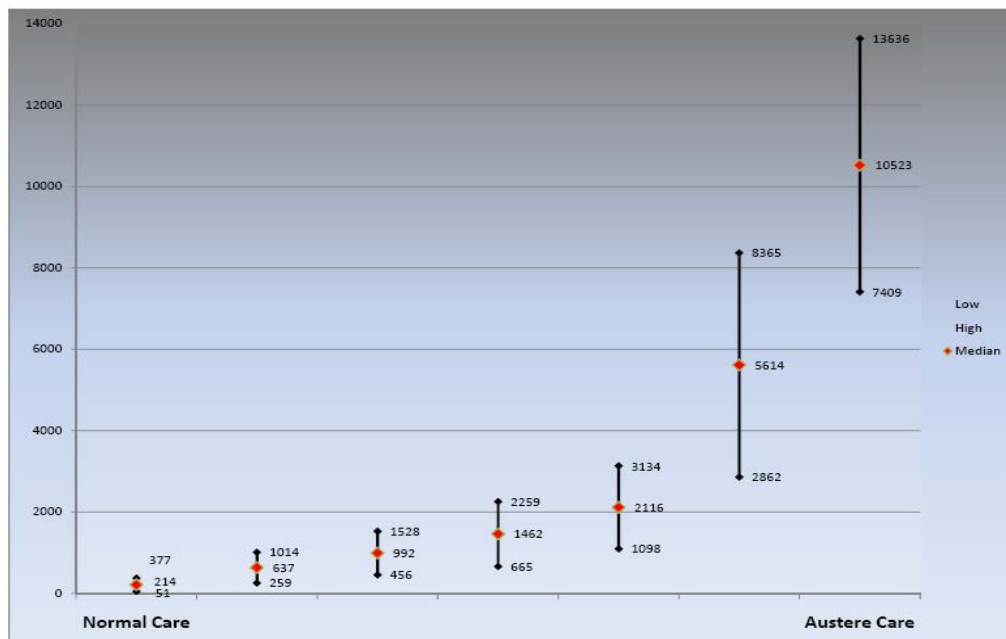
To assess the capacity of RITN, a survey was sent to all centers asking how many casualties could be received based on a various levels of care provided, ranging from standard operations to crisis standards of care. The results were surprising since the maximum number of casualties that could be received applying the most austere levels of care exceeded 13,000, a figure which exceeds the current capacity of the complete NMDP Network (Figures 13 and 14). The high austere capacity levels reflect the innovative care plans envisioned by the RITN centers to respond in the event of a catastrophic event.

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Figure 13. RITN Capacity Survey (2011)

#	Question	Low Estimate	High Estimate
1	How many patients could you receive in your existing BMT unit with no changes (e.g., no early discharges/transfers, no delayed admissions, no addition of beds, etc...)?	51	377
2	How many patients could you receive now in your existing BMT unit with modest changes (e.g., early discharges/transfers, a few delayed admissions, addition of beds from Hem/Onc service, etc...)?	259	1,014
3	How many patients could you receive now in your existing BMT unit with aggressive changes (e.g., aggressive discharges/transfers, many delayed admissions)?	456	1,528
4	How many patients could you receive now with spill-over into other areas of your hospital (Hem/Onc, med/surg, ICU), assuming no alterations in standards of care?	665	2,259
5	How many patients could you receive now in your existing BMT unit with aggressive changes and spill-over into other areas of your hospital (Hem/Onc, med/surg, ICU), assuming some alterations in standards of care?	1,098	3,134
6	How many patients could you receive now with the above and utilizing additional hospitals in your community?	2,862	8,365
7	How many patients could you receive now with the above and incorporating large austere emergency treatment facilities that have been previously planned for (e.g. pre-defined: dormitories, gymnasiums, domed stadiums, and assuming major alterations in standards of care)?	7,409	13,636

Figure 14. RITN Capacity Survey (2011)



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Aim A.2.2: Develop and test standard operating procedures, in conjunction with core transplant centers, to manage the activities required to HLA type siblings of casualties to evaluate their potential as Hematopoietic Stem Cell (HSC) donors for their affected family member.

The focus of this Aim was to develop and test standard operating procedures that would manage the activities required to HLA type and evaluate transplant suitability of related donors (siblings) of casualties.

In the event of a mass casualty incident resulting in marrow toxic injuries, such as an improvised nuclear device, there could be thousands of victims that will have suppressed immune function exhibited through the hematopoietic system. Some of these victims may require a transplant; those that fall into this category will need to have potential related donors HLA typed quickly. Since the country will likely be in turmoil, it may be difficult to bring these related donors to the transplant center for typing or collection. The NMDP's existing processes and laboratory contracts are ideal for providing a means to fill this need.

During this period of performance, the manual related typing process procedure was revised and updated. This process could be implemented to type of thousands of relatives of casualties in an emergency situation.

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II.A. Contingency Preparedness – Hypothesis 3:

NMDP's critical information technology infrastructure must remain operational during contingency situations that directly affect the Coordinating Center.

Aim A.3.1: Disaster Recovery: Ensure NMDP's ability to access and utilize its information management and communication infrastructure in a contingency situation in which its Minneapolis Coordinating Center is damaged or destroyed.

A Disaster Recovery exercise of Tier 1 applications was successfully conducted the first week of October 2010 with a Recovery Time of 42 hours.

Tier 1 Comprised of the following Core Operations Critical Applications and supporting Systems with a Recovery Time Objective (RTO) of 48 hours:

- STAR® 2 System
- TRAXIS®
- STAR Link®
- SEARCH Link™
- CORD Link®
- CRIS Link®
- FormsNet™ 1 & 2
- KEY Link
- Webmail
- Mail Services
- Network Website
- B2B
- ESB

Factors that contributed to the success of this Disaster Recovery Test:

- Used the Disaster Recovery “Warm Site” which provides the opportunity to minimize efforts for recovering from a real disaster
- Used NetApps Filer Mirrors for Database restorations which provides near real-time data without the use of tape restores
- Updated and validated documentation supporting new technologies implemented in Disaster Recovery
 - Example: Enterprise Service Bus (ESB) implemented in the past year was tested
- Utilized the Quality Assurance testing team to develop explicit test procedures as first line of validation
- Conducted a thorough system and application integration tests with support from development staff

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Aim A.3.2: Operational Continuity Planning:

The focus of this Aim is to improve organizational resiliency to severe operational disruptions through Operational Continuity Planning. In the event that the Coordinating Center is not available for an extended period of time, critical tasks will have to be conducted at an alternate location. To meet these needs, the NMDP has an Operational Continuity Plan (OCP), including a Critical Task List to prioritize the response.

The cohesive and efficient operation of the NMDP Network is dependent upon the availability of staff and systems at the Coordinating Center, as well as their resiliency to incidents that negatively impact operations. To help ensure that NMDP operations are able to continue despite operational interruptions, an OCP is formally established to mitigate impact from catastrophic incidents. This plan ensures that critical operations continue, or are resumed as quickly as possible, in the event of operational interruptions.

The NMDP's formal OCP is maintained by the Operational Continuity Planner. The Operational Continuity Planner ensures the proper focus is placed on this important area of operations resiliency and recovery. The Operational Continuity Planner works with each operational unit to determine priorities of work and recovery to accomplish their critical tasks. The planner then works with the Information Technology Disaster Recovery team to incorporate these operational unit needs into the disaster recovery plans where feasible. Appendices to this plan include the Critical Task List (a prioritized list of essential tasks to continue operations) and the Critical Document Register (a list of essential electronic documents provided by each department and the corresponding location on the network).

During this period of performance, the second NMDP Operational Continuity Exercise was conducted. The purpose of the exercise was to validate the ability of selected department staff to execute identified departmental critical tasks from a remote location; in this case, the remote location was a Radisson hotel conference room. This test involved seven core operational units: Donor Resources, Search and Transplant, Legal, Risk, and Network Affairs, Patient Services, Quality Systems, Be the Match Foundation, and Scientific Services. The exercise successfully validated the ability to establish remote data center connectivity and unified communications at an ad hoc Critical Staff Recovery Site, successfully assessed the capability of seven key department staff liaisons to complete identified critical operational tasks from this alternate locations, and captured shortcomings for corrective action.

To support the establishment of multiple Critical Staff Recovery Sites, long lead time equipment, which is necessary for a group of staff to work from the same location, was purchased, including virtual private network connection devices, routers, and firewalls. Additionally, 50 cellular network broadband modems were acquired to be used by staff to connect to the internet and perform their duties in the event that there is not enough space initially at a Critical Staff Recovery Site.

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The Operational Continuity Planner works closely with the Information Systems Disaster Recovery team to ensure that operational unit needs are taken into consideration for Information Services (IS) disaster planning. This includes coordination of priority of system recovery to holding a gap analysis review of the OCP and the disaster recovery plan. This review walked through an incident that would impact NMDP operations and, at critical stages, the group discussed what each would be working on and what resources would be required to meet the organizations needs. This resulted in a list of key gaps that were prioritized for the following year for funding and implementation.

Effective communications are essential when responding to any disaster, emergency, or operational interruption. The NMDP is authorized to distribute, through support of the National Communications System, over 150 Governmental Emergency Telecommunications Service (GETS) emergency calling cards. These emergency calling cards allow users to place phone calls during times of significant telephone network congestion. Quarterly user tests were conducted to ensure GETS card accountability and the users' ability to successfully place calls.

Satellite telephones are part of the emergency communications program as a means of last resort to ensure communications during a disaster that impacts landline telecommunications equipment in the United States. Most of these portable satellite telephones were issued to RITN centers, and the remaining phones are maintained at the NMDP Coordinating Center for use during a disaster.

The Operational Continuity Planner conducted site visits to multiple NMDP operated donor centers to review the NMDP Operational Continuity Action Guide with each site manager and staff. During these visits, the guide was reviewed with each manager to ensure they know what to do in the event of the following major hazards, if applicable to their location, including:

- Hurricane
- Flooding
- Earthquake
- Tornado
- Winter storm
- Power outage
- Chemical spills
- Extreme heat

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II.B. Rapid Identification of Matched Donors – Hypothesis 1:

Increasing the resolution and quality of the HLA testing of volunteers on the registry will speed donor selection.

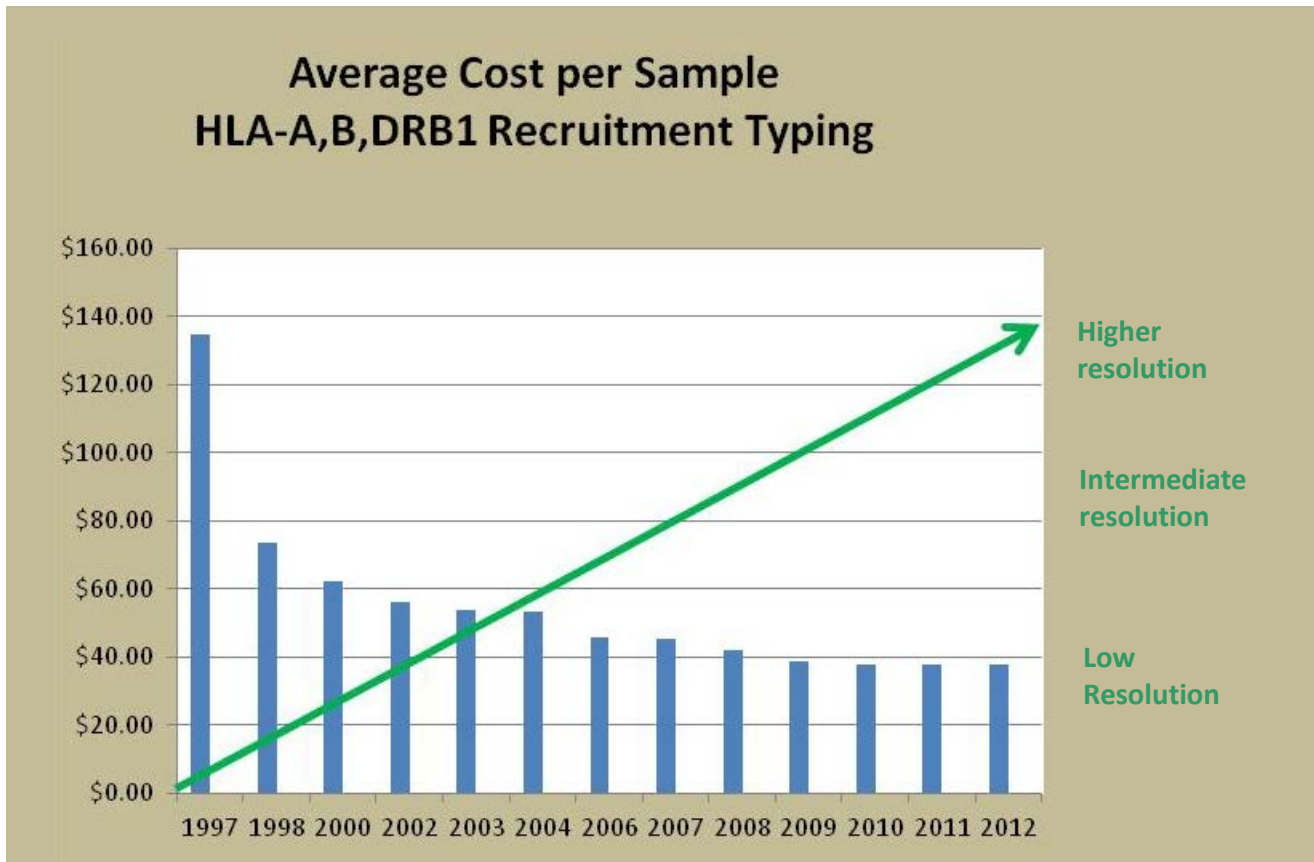
Continued advances in laboratory methods and supporting equipment have positively impacted the level of typing resolution for newly recruited volunteer donors. Six of the laboratories reported results at higher than intermediate level of resolution and one laboratory reported intermediate resolution. Two laboratories reported HLA-A, B, C and DRB1, while five laboratories reported HLA-A, B and DRB1. Of all newly recruited donors, 45% were typed at HLA-A, B, C and DRB1.

In January, 2010, the NMDP began selecting younger newly recruited donors to receive HLA-C typing in addition to HLA-A, B and DRB1. Between January 2010 and May 2010, all males 18-40 years of age were typed at HLA-C. In May, 2010, an additional laboratory was added to provide HLA-C typing and females 18-40 years of age were included in this selection when capacity was available. As of July, 2010, seven laboratories provided HLA typing. From 01/05/2010 through 09/27/2010, 120,105 young donors were typed at HLA-A, B, C, and DRB1 at the time of recruitment.

Over the past 15 years, the NMDP has successfully reduced the cost of HLA typing by over 70% while increasing the resolution and quality (Figure 15). The vision and efforts of the Navy to continually press the HLA community in this direction and to lead the advancement and development of new typing technologies has been instrumental in achieving gains in resolution and decreases in cost.

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Figure 15. Per sample cost for new donor recruitment typing has decreased over 70% and the resolution has continued to increase since 1997.



If a patient does not find a matched donor and is in urgent need, patient-focused drives can be held and the donor registration process can be expedited, shortening the length of time to listing from 6-8 weeks to 3 weeks. This process includes time to enter demographic data, confirm financial coverage, ship and receive the samples, and complete the HLA typing. Demographic data are entered within 72 hours for expedited samples and they are shipped the next scheduled day, Monday through Thursday. In case of a contingency event, high volumes of samples could be processed and shipped quickly using this established process.

The NMDP's exacting quality control processes have successfully increased the quality of typing received through the contract laboratory network. The method of inserting blind quality control samples into each laboratory's shipment of volunteer donor samples has provided more than 12 years of data tracking the accuracy of high volume typing. Over this time, the accuracy rates have continued to improve, as documented by decreased monthly error rates and decreased discrepancies as donors are selected for patients and retyped by other laboratories. The effectiveness of this program and the efforts of a highly qualified high-volume HLA typing

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laboratory network has resulted in a combined HLA class I and class II QC accuracy rate, from Oct 2009 to Sep 2010, of 99.9%.

Aim B.1.1: Increase Registry Diversity

Expand the genetic diversity of the Registry through continued addition of adult donors and cord blood units, utilizing high volume HLA typing methodologies.

During NMDP's FY10 (10/01/2009-09/30/2010), NMDP donor centers (including DoD) and recruitment groups recruited 270,284 minority race and 305,624 Caucasian donors, which were typed at minimum for HLA-A, B and DRB1. Navy funding contributed to the addition of 145,553 NMDP recruited donors (excluding DoD). This added a culturally diverse group of new donors to the Registry.

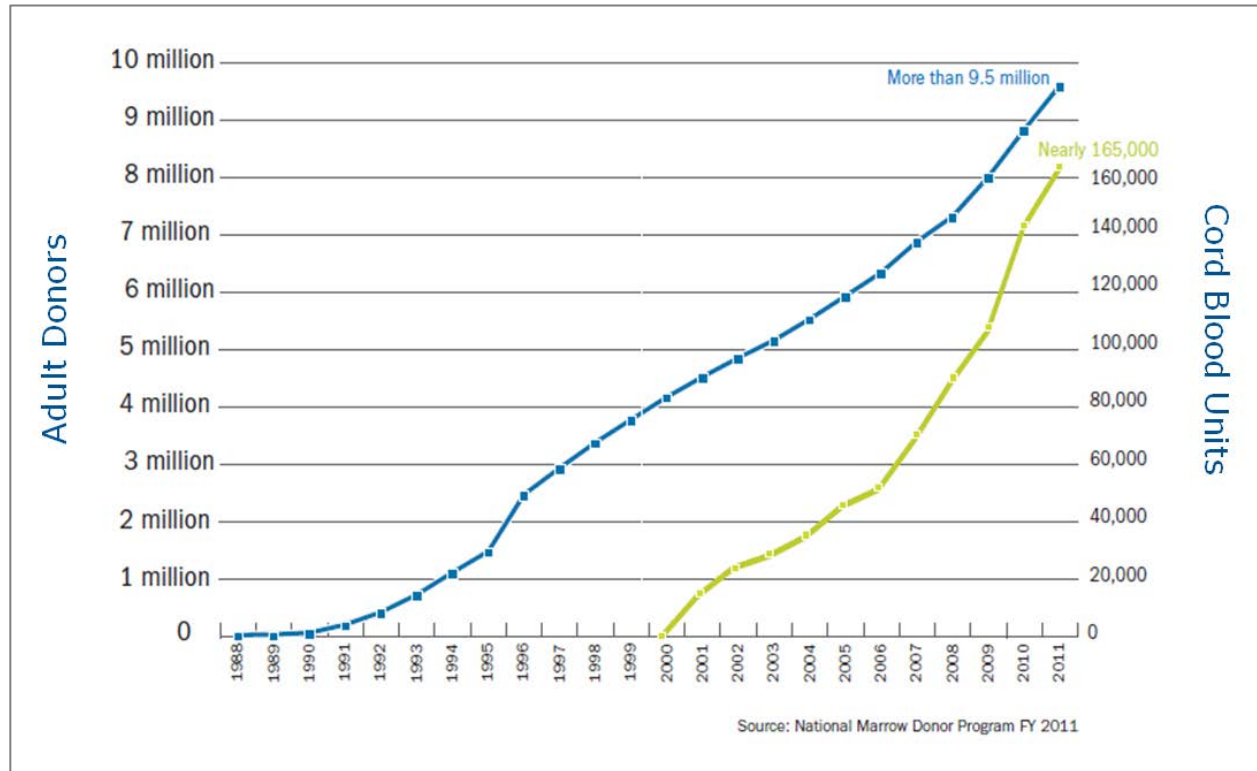
Advancing technology improved performance and pricing

Advances in laboratory methods and technology continue to have a positive impact on lab performance and pricing:

- 96% of new donors received higher than intermediate HLA-A, B typing
- 45% of new donors received intermediate HLA-C typing
- 100% of new donors received higher than intermediate HLA- DRB1 typing
- Blinded quality control testing accuracy rate was 99.88%, exceeding the project requirement of $\leq 2.0\%$.
- On-time testing completion rate was 98.8%, meeting the project requirement of a minimum of 90.0% of typing results reported within 14 days of shipment of samples.
- All new donors were typed at HLA-A, B, and DRB1. 34% of new donors were also typed at HLA-C. All typing was at intermediate resolution.
- The cost of HLA typing continues to decrease as technology improves; during the period March 2010 through August 2010 the average price per sample was approximately \$37.60, compared to \$134.75 in 1997, which represents a decrease of over 70%.

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Figure 16. Total Growth of the Be The Match Registry: Adult Donors and Cord Blood Units



HLA Quality Testing

Two HLA typing projects were completed during this contract period. These studies were designed to increase the resolution and quality of HLA typing on the registry to potentially speed donor selection and correctly characterize the match for searching patients, especially from diverse populations.

The first project evaluated the accuracy of DRB1*16:08 reported in the NMDP Registry. The allele was described in 1996, but had not been reported on an adult volunteer sample in the NMDP Registry since 2001. 197 donors with DRB1*16:08 reported and with a stored sample at the NMDP repository were retyped at intermediate resolution DRB1 to determine if these samples truly carried DRB1*16:08. 100% of the samples came back as a different DRB1 allele, (Table 1). A poster abstract detailing the results of the retyping project for DRB1*16:08 was presented at the 2012 European Federation of Immunogenetics (EFI) meeting.²

Table 1: Results of DRB1*16:08 retyping project.

DRB1*15:01	DRB1*15:02	DRB1*15:03	DRB1*15:22	DRB1*16:01	Indeterminate
151	19	2	1	21	4

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The NMDP maintains lists of rare alleles as a service to the American Society for Histocompatibility & Immunogenetics (ASHI). These lists are derived from HLA allele-level typings of patients, adult volunteers, and cord blood units in the NMDP Registry. Careful review of the rare alleles reported to the NMDP on adult volunteer samples revealed results that were suspicious and may have been incorrectly reported due to various reasons including:

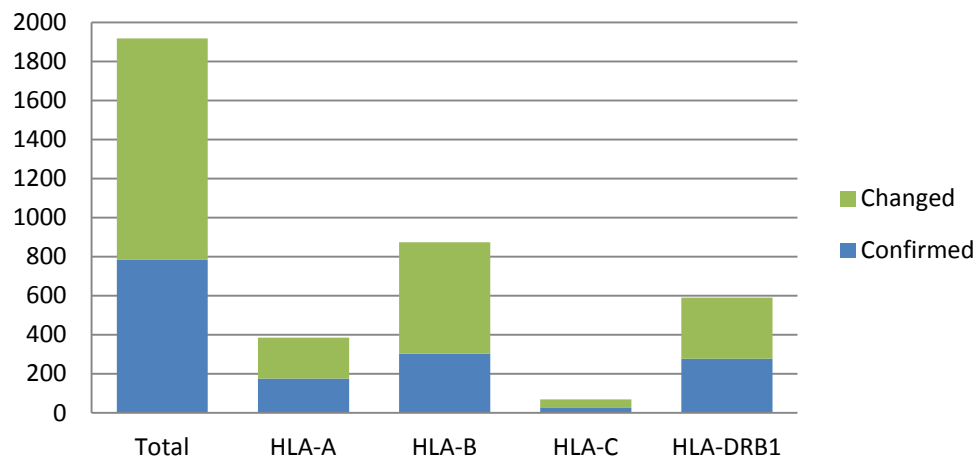
- Typing methodologies used to test the rare allele were problematic, resulting in a correction of some of the rare allele results
- Rare allele was typed more than 4 years ago and the allele has not been reported since
- Presence of two rare alleles in a donor typing
- Primary data interpretation didn't match the rare allele reported
- Rare allele was reported more than once on the same day with no haplotype to indicate the donors are possibly related

Samples were identified using the above criteria and retyped primarily by sequence based typing (SBT) methodologies to confirm the allele in question. The results of this retyping project are summarized in Table 2 and Figure 17.

Table 2 : Summary of the results of the rare allele typing project

	samples typed	Confirmed	% confirmed	Changed	% changed
Total	1919	784	40.9%	1135	59.1%
HLA-A	386	175	45.3%	211	54.7%
HLA-B	874	304	34.8%	570	65.2%
HLA-C	69	28	40.6%	41	59.4%
HLA-DRB1	590	277	46.9%	313	53.1%

Figure 17: Rare alleles confirmed or updated on the NMDP Registry.



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An abstract summarizing these data was presented at the 2010 ASHI meeting and at the 2011 EFI meeting.^{3,4}

Results of these retyping projects improved the HLA typing accuracy and quality of listed adult volunteers. These projects highlight the importance of continuous monitoring of the Registry data and the necessity to upgrade typing routinely in order to provide the most accurate HLA data for searching patients.

Aim B.1.2: Evaluate HLA-DRB1 High Res Typing

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.1.3: Evaluate HLA-C Typing of Donors

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.1.4: Evaluate Suitability of Buccal Swabs

The 5-year Sample Storage Research Study began in September 2007. Samples from 30 fully HLA characterized volunteer quality control donors were collected, processed, and stored at the NMDP Repository. The samples, which consisted of fresh blood, blood spotted onto Whatman[®] 903 filter paper and buccal swabs for each donor, were sent to two laboratories in September 2007 to initiate the study (Time Point Zero). One laboratory was contracted to perform high resolution typing for HLA-A, B, C, DRB1, and DQB1. A second laboratory was contracted to perform intermediate resolution typing for HLA-A, B, C, and DRB1 and also to evaluate the quantity and quality of DNA within each sample type. Complete results were received from each of the two laboratories. All typing results were 100% accurate, and the evaluation of the DNA was complete and thorough. The results obtained at Time Point Zero are used as the baseline for comparison and evaluation of the stability and usefulness of the DNA stored in each sample type for the next 5 years. Results from this study will provide key quality parameters for NMDP operational decisions concerning sample storage and may also contribute sample storage guidelines for other registries.

In September 2011, stored donor samples were sent to the laboratories for evaluation at the 4 Year Time Point. All HLA results were 100% accurate. One measure of the DNA quality derived from the stored samples is the frequency of repeated HLA testing for each locus.

- The laboratory performing intermediate resolution HLA-A, B, C, and DRB1 reported zero repeats.
- The laboratory performing high resolution HLA-A, B, C, DRB1, DQB1 reported the following repeats (see Table 3 below):

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Table 3. Summary of repeat testing required by sample type for the Sample Storage Research Study.

Sample Type*		Whole Blood		Filter Paper		Swab	
Time Point	Resolution	# Repeat loci / Total # loci	%	# Repeat loci / Total # loci	%	# Repeat loci / Total # loci	%
Zero	High	3/150	2.0%	2/150	1.3%	2/150	1.3%
One Year	High	0/150	0.0%	0/150	0.0%	0/150	0.0%
Two Year	High	2/150	1.3%	1/150	0.7%	4/150	2.7%
Three Year	High	0/150	0.0%	0/150	0.0%	5/150	3.3%
Four Year	High	0/150	0.0%	0/150	0.0%	5/150	3.3%

*30 samples/sample type * 5 loci = 150 loci/sample type

The data shows that all sample types remain stable for obtaining HLA results, with limited repeat testing. The 6 laboratories currently performing HLA typing on new donor recruitment samples report a donor sample repeat rate between 3-5 % of loci tested for buccal swab samples received each week. The results from Time Point 4 are within this same range.


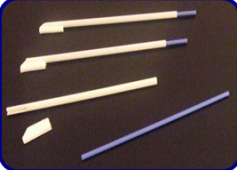
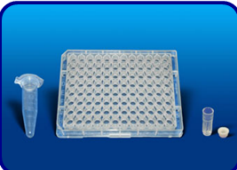
Alternate Sample Collection Methods Study

A limited feasibility study was conducted to evaluate alternate sample collection and storage methods, including possibilities for storage formats that offer increased sample lifetime, more compact storage, and greater downstream sample utility for further detailed typing. The potential to store DNA in a stable form at room temperature is an attractive possibility for the long-term storage of a resource that would be renewable and in an intact state for typing after decades of storage, when needed for patient or contingency needs. Sample sets were collected from 15 volunteer donors for evaluation. Each donor collected samples using:

- Oragene® DNA saliva sample collection kit from DNA Genotek
- CEP Swab®, an ejectable-tip buccal swab from Fitzco, Inc.
- Standard NMDP cotton-tipped buccal swab on polystyrene shaft

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Figure 18: Summary of technologies for comparison to NMDP cotton-tipped buccal swab

	Oragene® saliva collection kit <ul style="list-style-type: none">• DNA Genotek, OG-500• Claimed advantages: DNA stability at room temperature; higher quantity of DNA
	CEP-Swab®, ejectable-tip buccal swab <ul style="list-style-type: none">• Cotton-based fibrous material in a matrix format, from Fitzco, Inc.• Claimed advantages: Compact storage of tip only; ease of use for laboratory staff
	GenTegra® tubes for DNA storage <ul style="list-style-type: none">• GenVault screw-cap tubes for long-term dry storage of purified DNA (GTD2025-S)• Claimed advantages: DNA stability at room temperature; very compact storage

- Samples were sent to 3 laboratories (5 sample sets per laboratory) for analysis of the following attributes:
 - Quality and quantity of DNA
 - High resolution typing at HLA-A, B, C, DRB1, DQB1, and DPB1
 - Extraction and processing of DNA for dry storage in GenTegra tubes from GenVault
 - Report on the advantages and disadvantages of each sample type and storage method, from a laboratory perspective

All collection methods produced good quality DNA for HLA typing from fresh samples, with all labs obtaining high resolution typing results at all loci that are consistent with NMDP results on record. Results from each lab are summarized below.

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Figure 19: Summary of Lab Results for Alternate Sample Collection Methods Study

LAB A	Oragene Saliva	CEP Swab	Cotton Swab
Starting material	500 µl	1 swab	1 swab
DNA yield (ng/µl)	114.05 avg (42.3 – 241.4)	24.79 avg (9.5 – 60.5)	30.16 avg (12 – 50.8)
260/280	1.84 avg	2.04 avg	1.94 avg
HLA typing	All reported results consistent with NMDP high resolution typing on record		

LAB B	Oragene Saliva	CEP Swab	Cotton Swab
Starting material	100 µl	1 swab	1 swab
DNA yield (ng/µl)	330 avg (28 – 1140)	197 avg (40 – 452)	147 avg (84 – 240)
260/280	1.87 avg	1.06 avg	1.09 avg
HLA typing	All reported results consistent with NMDP high resolution typing on record		

LAB C	Oragene Saliva	CEP Swab	Cotton Swab
Starting material	Volume not reported	1 swab	1 swab
DNA yield (ng/µl)	218.71 avg (137.25 – 332.12)	2.89 avg (1.34 – 5.14)	19.89 avg (12.72 – 28.46)
260/280	1.81 avg	1.42 avg	1.86 avg
HLA typing	All reported results consistent with NMDP high resolution typing on record		

Comments from the laboratories on each method were gathered and are summarized below in Figure 20.

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Figure 20: Lab Comments on collection method for Alternate Sample Collection Methods Study

Oragene Saliva Sample	<ul style="list-style-type: none"> •+ Highest DNA yield; abundant sample •+/- High quality DNA, but requires purification step • - High viscosity of sample is problematic for pipetting • - Transfer to new tube for testing risks cross-contamination • - Time consuming and tedious extraction procedure; requires vigorous pipetting and vortexing to dissolve DNA pellet • - Not optimized for high through-put; requires centrifugation
CEP-Swab	<ul style="list-style-type: none"> •+ Convenient to eject tip • - Lower DNA yield than cotton swab in 1 lab • - High absorbency of tip makes processing difficult • - Tip is too tall to fit plates well
Cotton Swab	<ul style="list-style-type: none"> •+ Simple and convenient handling for high through-put • - Manual snipping of shaft

Comments on the donor user experience were gathered and are summarized below.

Figure 21: Donor User Experience: Alternate Sample Collection Methods Study

Donor Comments	Oragene Saliva	CEP-Swab	Cotton Swab
Average Satisfaction Score (1-10, with 10 = Very Satisfied)	3.6	6.5	9.0
Positive comments	•Easy to use	•None reported	•Easiest to use •Quick
Negative comments	<ul style="list-style-type: none"> •Unappealing to spit into the tube, especially in front of other people •Time consuming (up to 10 min) •Difficult to tell when fill line is met (bubbles) •Difficult to generate enough saliva •Caused mild nausea •Complicated 	<ul style="list-style-type: none"> •Uncomfortable shape and material (some cases of bleeding) •Swab disconnected from shaft during swabbing •Disintegration during swabbing 	•None reported

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Conclusions:

- All collection methods produce good quality DNA for HLA typing from fresh samples
- The disadvantages of the CEP-Swab, from both the donor and laboratory perspectives, outweigh any potential advantages
- Optimization of Oragene saliva sample methods for high-throughput handling would need development; abundant high-quality DNA was obtained from this sample type, but DNA purification steps were cumbersome and time-consuming for laboratory staff

Future studies on stored samples will provide more information. Duplicate samples and extracted, dry-state DNA from each of the sample collection methods were stored, for future evaluation.

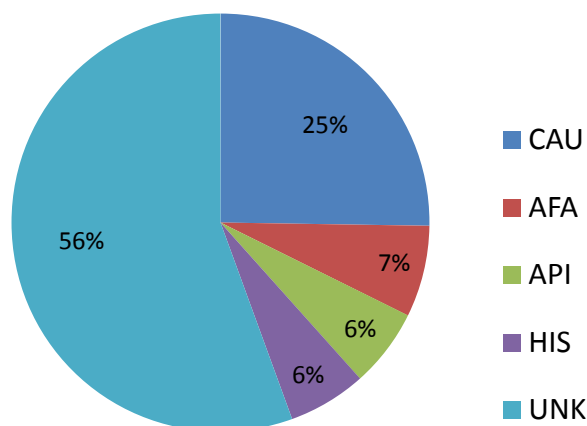
Aim B.1.5: Enhancing HLA Data for Selected Donors

A study designed to evaluate the benefit of adding HLA-DRB1 to selected HLA-A, B only typed donors was completed during this award period. HLA-A, B only typed adult donors comprise 12% of the NMDP's Be The Match Registry® and are infrequently used for searching patients. This study was designed to evaluate:

- Whether testing HLA-A, B only donors for patients with no potential 6/6 allele matched donors is a good allocation of patient resources
- If testing HLA-A, B only donors adds genetic diversity to the registry, presenting potential options for the pool of current and future searching patients

A total of 210 patients with no potential 6/6 allele matched donors on their preliminary search list were identified for evaluation. The race/ethnicity distribution of the cohort is shown in Figure 22.

Figure 22: AB only study patient race/ethnicity (n=210)



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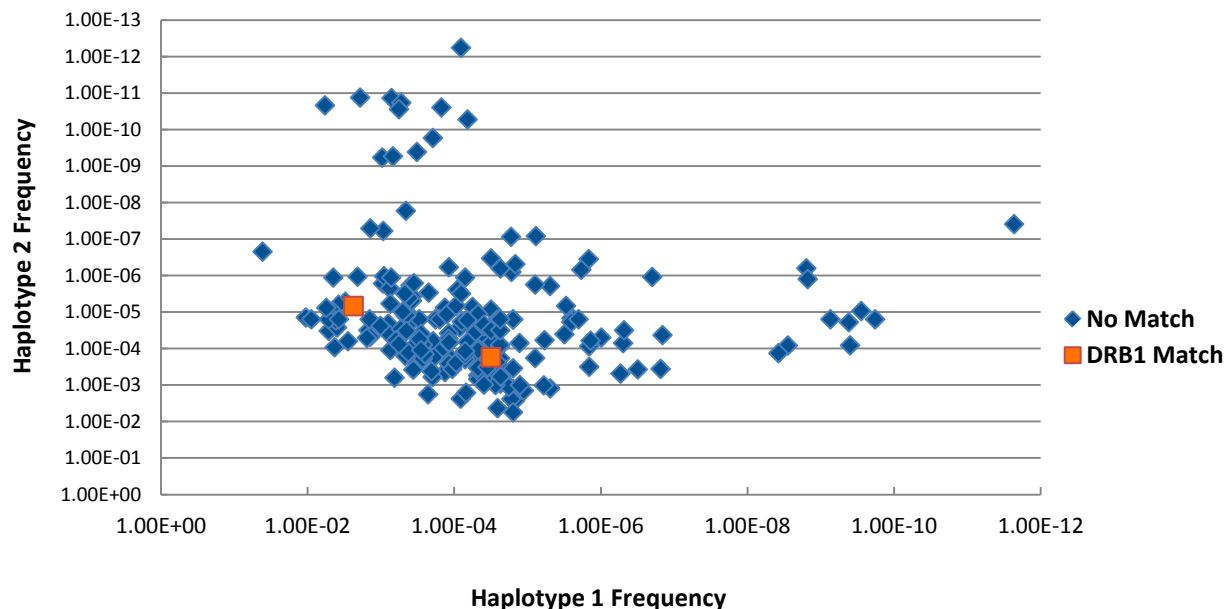
All HLA-A, B only donors with repository samples that were potential matches for the 210 study patients were typed at DRB1; the median number tested per patient was 4, with a range of 1 to 231 HLA-A, B only donors typed. Donor DRB1 typing results were compared to the corresponding patient's DRB1 alleles to confirm matching status.

Of the 3,735 donors typed for the 210 patients, only two matched their respective patients at DRB1.

- One DRB1 matched donor was ultimately an 8/10 (5/6), with A and C allele mismatches to the respective patient
- The other DRB1 matched donor went to transplant for the corresponding patient
 - 10/10 match
 - Carried an A/B/DRB1 phenotype not previously identified on the registry

Both matches occurred for patients with moderate individual haplotype frequencies (Figure 23).

Figure 23: Estimate Patient Haplotype Frequency



These results showed limited benefit from HLA-A, B only donor testing for patients considered in the study, since DRB1 matched donors were identified for only 2 of the 210 patients evaluated.

Other considerations when testing HLA-A, B only donors:

- Most HLA-A, B only donors were typed by serology leading to additional underlying HLA disparity at HLA-A, B and C.

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- Only donors with stored DNA samples were tested, representing only 26% of the total HLA-A, B only donors on the 210 patient searches
 - Additional DRB1 matches may have been found if every AB only donor for each patient had been tested
 - Study patients represented challenging searches, with no potential 6/6 allele matched donors
 - A different success rate may be determined if selecting patients with ≥ 1 existing A/B/DRB1 matched donor, implying a more frequent HLA phenotype

In conclusion, while the probability of finding a DRB1 matched donor from the HLA-A, B only pool is likely very low, the possibility remains, even with an uncommon patient A/B/DRB1 phenotype. Based on the low rate of success, the strategy of testing HLA-A, B only donors may be best used in conjunction with selection of the best potential mismatched donor to avoid delaying transplant.

The above information was presented as a poster abstract at the 2012 ASBMT Tandem Meeting.⁵

As evidenced in the Figure 4 above, a small proportion (19%) of patients in the AB only DRB1 typing project were known to be non-Cau. To further assess the second goal of the study, which was to test whether adding HLA-DRB1 to HLA-A, B only typed donors adds diversity to the overall registry of 10 million adult donor, all minority AB only donors with a self described broad race/ethnic groups of AFA, API, CAU, or HIS with a stored repository sample (n=729) were identified for DRB1 typing and phenotype evaluation. The CAU group carried specific detailed race designation of Mediterranean, Mideast, and/or North Coast of Africa (MENAF).C).

In all race groups, over 25% of the donors carried an uncommon or unique phenotype contributing additional diversity to the registry (Table 4).

Table 4: Resulting ABDRB1 Phenotype Commonality by Race (n=729)

Race	Total	Unique Phenotype	Uncommon Phenotype	Total % Unique or Uncommon	# of ABDR Donors on Registry
AFA	48	19 (40%)	11 (23%)	63%	680,000
API	58	8 (14%)	16 (28%)	41%	707,000
HIS	79	15 (19%)	5 (6%)	25%	470,000
MENAF	544	77 (14%)	74 (14%)	28%	80,000
Total	729	119 (16%)	106 (15%)	31%	1,937,000

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These results show that HLA-A, B only donors can have significant diversity and typing the remaining 64,000 minority HLA-A, B only donors is a cost efficient way to expand the HLA diversity of the registry with unique or uncommon phenotypes. Interestingly, five donors in this cohort were requested for additional typing by a searching patient in the subsequent months after DRB1 typing was completed.

The above information was accepted for poster presentation at the 2012 ASHI meeting.⁶

Aim B.1.6: Maintain a comprehensive quality control program

No funding was requested under this Aim for the 0204 budget cycle.

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II.B. Rapid Identification of Matched Donors – Hypothesis 2:

Primary DNA typing data can be used within the registry to improve the quality and resolution of volunteer donor HLA assignments.

Aim B.2.1: Collection of Primary Data

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.2.2: Validation of Logic of Primary Data

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.2.3: Reinterpretation of Primary Data

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.2.4: Genotype Lists & Matching Algorithm

The focus of the work under this aim was to develop data standards and methods to allow the reporting and storage of primary DNA sequences and to convert this data into up-to-date genotype lists for use in the matching algorithm. As the technology for HLA typing has shifted from oligo-probe based methods to sequencing based methods, the system needs to be modified to adapt to this change.

In an effort to promote the use of electronic data standards for reporting data to NMDP and others within the field, a comprehensive data dictionary document was developed that offers an overview and comparison of several different versions of the electronic messaging format HML (Histo-Immunogenetics Markup Language):

[http://bioinformatics.nmdp.org/HLA/HLA_Typing/HML/HML_Data_Dictionary_\(PDF\).aspx](http://bioinformatics.nmdp.org/HLA/HLA_Typing/HML/HML_Data_Dictionary_(PDF).aspx).

The latest version of the HML message format, v0.3.3, has been implemented to support SBT and in particular the use of SBT as an ambiguity resolution technique:

http://bioinformatics.nmdp.org/HLA/HLA_Typing/HML/HML_0_3_3/HML_Version_0_3_3.aspx). This work was presented as an abstract at the 2010 ASHI meeting. As of April 2011, the NMDP has implemented support for SBT HLA primary data collection in its operational systems. At this time, several changes were also implemented to support the new, second phase of the transition to the new HLA nomenclature that began in April 2010.

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II.B. Rapid Identification of Matched Donors – Hypothesis 3:

Registry data on HLA allele and haplotype frequencies and on the nuances of HLA typing can be used to design computer algorithms to predict the best matched donor.

Aim B.3.1: Phase I of Expectation Maximization (EM) Haplotype Logic

The focus under this aim has been to perform the research and development necessary to deliver a new implementation of the HLA matching algorithm (HapLogicSM) that can compute the probability that the donor typing will match the recipient at high-resolution across 5 HLA loci (HLA-A, B, C, DRB1 and DQB1) in order to better align, with clinical matching and speed, the process of identifying a matched donor.

As a pre-requisite to this work, the Bioinformatics team implemented a technology migration of the Search Server matching algorithm from the “C” programming language to Java in order to:

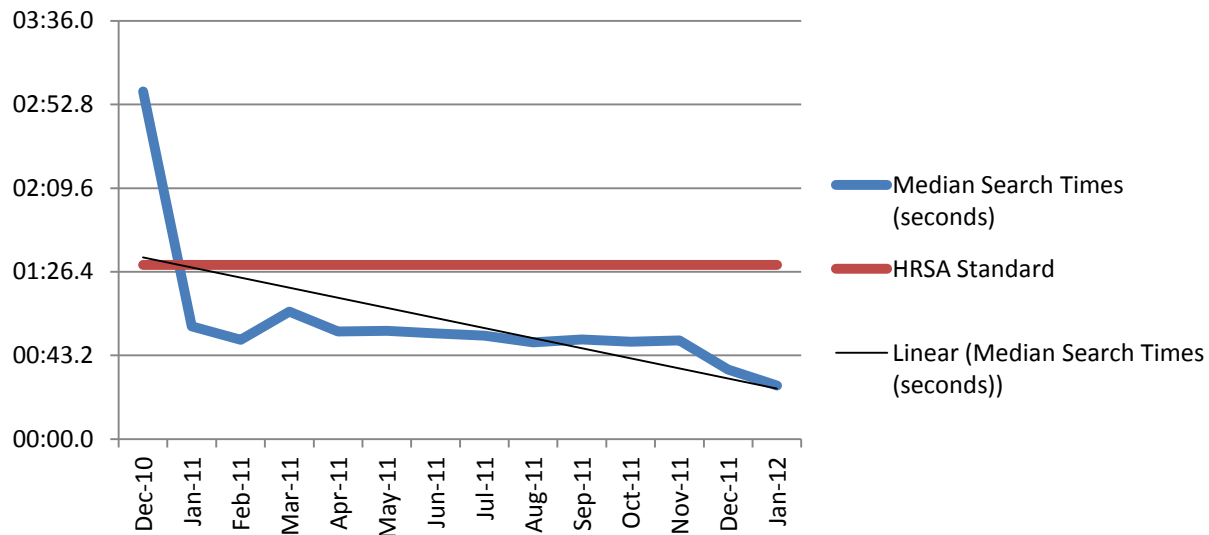
- Improve performance
- Increase flexibility for implementing and validating future algorithm changes
- Increase accessibility of the matching algorithm to dependent applications
- Reduce implementation redundancies

NMDP implemented the HapLogic algorithm with increased precision and clarity which included:

- 3 locus matching was increased to 5 locus matching
- x of 6 is now x of 8 or x of 10 predictions
- 5 broad race groups were expanded to 5 broad and 18 detailed race groups
- Ensuring visibility of NMDP’s best matched donors and CBUs
- More precision for mismatch searches
- Better alignment with clinical practice
- Realized performance improvements to the HapLogic algorithm, with a median search run time of 35 seconds (Figure 24)

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Figure 24: Median Search Time by Month (in seconds)



Additionally, as part of the re-platforming, a regression test suite was implemented with the algorithm in order to improve overall quality and increase confidence when embarking upon future changes. This foundation served as the basis for implementation of 10 of 10 HapLogic III matching predictions.

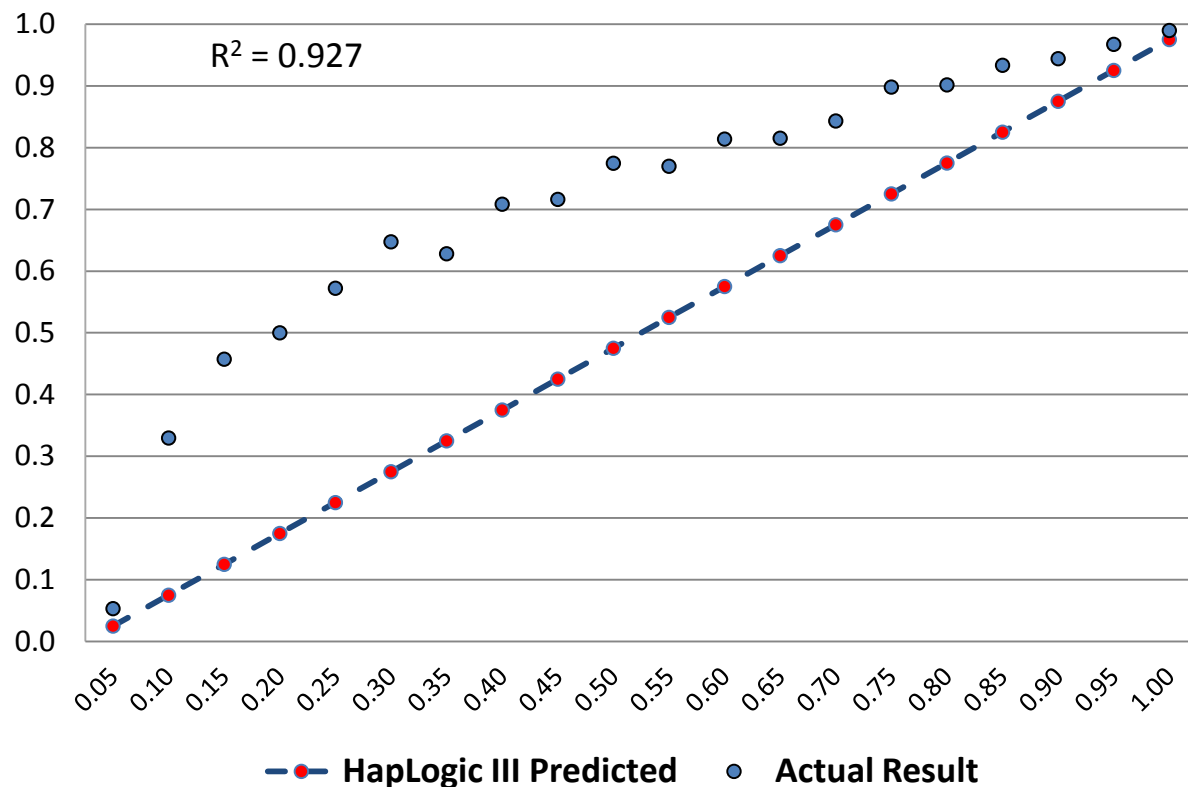
A prototype implementation of HapLogic was developed that allowed computation of 10/10, 9/10, etc. allele matching likelihoods as well as 8/8, 7/8, etc. This implementation represents the culmination of research conducted under several years of this research grant to develop haplotype frequencies for 21 distinct US populations at all 5 clinically relevant HLA loci. This effort is particularly noteworthy due to the fact that most donors in the registry are not typed at all 5 loci and almost all are reported with some degree of typing ambiguity (see aim IIB.3.2).

In order to evaluate the performance of the prototype matching algorithm, the Bioinformatics team compiled a validation dataset of over 58,000 patient-directed typing requests (2000-2008). The validation dataset was to be used to evaluate the accuracy, sensitivity/specificity, positive predictive value/negative predictive value for HapLogic for the 5 single-locus predictions (HLA-A, B, C, DRB1 and DQB1) and the x/10, x/8 and x/6 overall predictions. This same set was used to evaluate the previous HapLogic algorithm (which uses earlier HLA haplotype frequencies and has fewer outputs) to directly measure the improvement achieved with the enhanced algorithm and haplotype frequencies.

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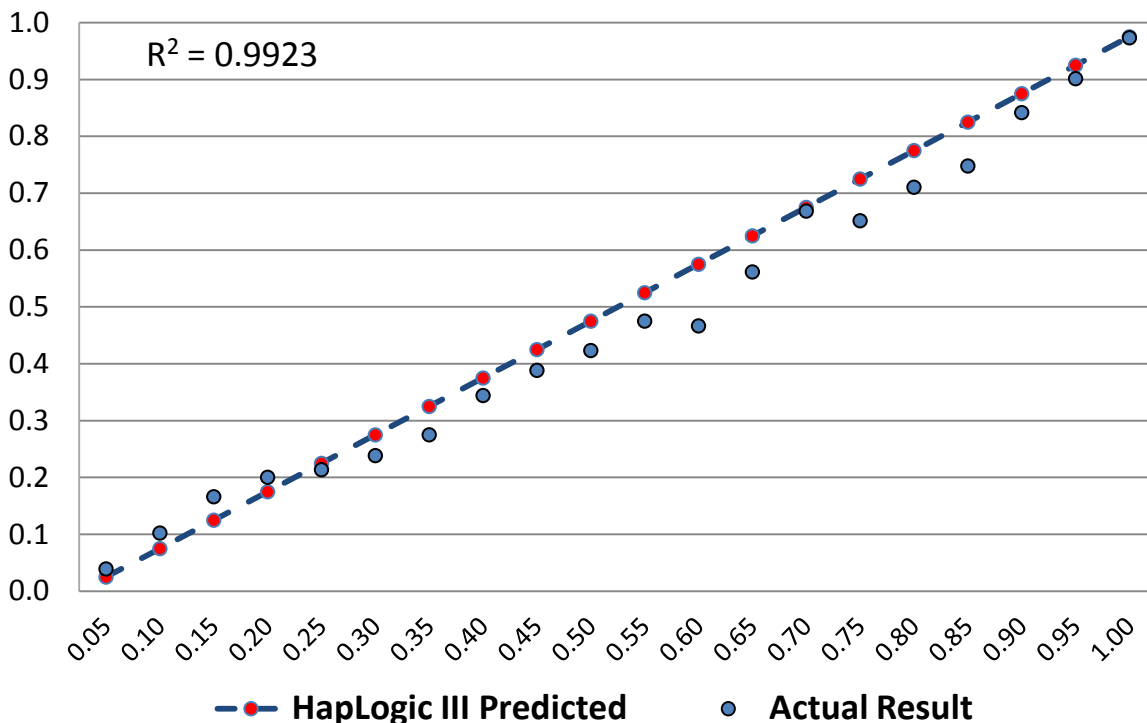
The performance of the algorithm agrees with the result of the typings, particularly when the algorithm provides a low (<0.1) or high (>0.9) prediction (Figure 25). The results in the 9/10 (single mismatch) cases correlate even better with expectation due to a number of factors, including the fact that there are more observations across the spectrum of likelihoods and more information about multiple haplotypes contributing to the overall likelihood of a mismatch (Figure 26). The performance of the algorithm was shown to be a substantial improvement over previous versions and so was transferred to the IT department for implementation during the next 6 months.

Figure 25: HapLogic Predictions: 10/10 Matches



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Figure 26: HapLogic Predictions: 9/10 Matches



Aim B.3.2: Enhancement of EM Algorithm

Six-locus high resolution HLA-A~C~B~DRB3/4/5~DRB1~DQB1 haplotype frequencies were calculated using all Be The Match™ Registry volunteer donors typed by DNA methods at recruitment. The developed method accepts mixed resolution HLA typing data into a modified EM algorithm. The full cohort consisted of 6.59 million subjects (donors and CBUs), of which only 25.8% were typed at the HLA-C locus, and 5.2% typed at the HLA-DQB1 locus, while all individuals were typed for HLA-A, B, DRB1.

One of the methods developed for dealing with typing ambiguity was to convert the primary DNA typing data stored over a period of 12 years (see Aim B.2.4) into genotype-lists for a more robust representation of the typing ambiguities.

A sub-analysis of 2.9 million subjects (donors and cord-blood units) was completed, with detailed race/ethnic information mapped to 21 population subgroups. Sample sizes at the detailed race level range from 1,242,890 for European Caucasian to 1,376 Alaskan Native or Aleut. These haplotype frequencies have been used to improve match predictions in the matching

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algorithm for hematopoietic stem cell transplantation (Aim B.3.1) and to improve the accuracy in modeling registry match rates (Aim B.3.3).

Table 5: 21 registry samples used to derived detailed race category haplotype frequencies

<u>Race_Code</u>	Description	Count	Typed_C	Typed_DQB1
AAFA	African American	300406	33079	6596
AFB	African	17094	913	309
AINDI	South Asian	133769	5365	1976
AISC	American Indian South or Centr	4289	254	101
ALANAM	Alaska Native or Aleut	1109	110	72
AMIND	North American Indian	30530	3891	1827
CARB	Black Caribbean	20609	2695	370
CARHIS	Caribbean Hispanic	82240	5580	1854
CARIBI	Caribbean Indian	3680	251	49
FILII	Filipino	36290	7013	850
HAWI	Hawaiian / Pacific Islander	7590	598	154
JAPI	Japanese	19185	1275	415
KORI	Korean	61561	3622	886
MENAF	MidEast/No. Coast of Africa	38492	4727	883
MSWHIS	Mexican or Chicano	212645	21784	7549
NAMER	North American	679521	82623	14618
NCHI	Chinese	68598	3835	1382
SCAHIS	South/Cntrl Amer. Hisp.	97769	8188	2321
SCAMB	Black South or Central America	3616	180	94
SCSEAI	Other Southeast Asian	18373	881	272
VIET	Vietnamese	17362	678	196

A first draft of a manuscript describing 6-locus haplotype frequency data has been circulated to co-authors. The goal is to publish this frequency studies along with the frequency tables.

Other haplotype frequency work that took place under this aim:

- Calculated Bone Marrow Donors Worldwide (BMDW) A~B~DRB1 haplotype frequencies by country. An abstract was accepted for the European Federation for Immunogenetics (EFI) meeting on these frequencies. Began processing of BMDW data in preparation for 5-locus BMDW haplotype frequency study.
- Completed a draft of a DPA1-DPB1 haplotype frequencies manuscript based on the Caucasian donors from the donor/recipient pairs typing project (Aim C.1.1) and submitted to the journal Immunogenetics.
- Initiated a project to automate quarterly haplotype frequency updates for periodic ongoing incorporation into and improvement of HapLogic.

Aim B.3.3: Optimal Registry Size Analysis

Under this aim, we have developed a set of tools and methods that use HLA haplotype frequency data to determine matching rates for the registry (adult donor and CBU) under a variety of

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growth scenarios. Summarizing work completed so far in applying the new haplotype frequencies (Aim B.3.2) to modeling the US population, we have created a first draft of the match rate results for a physician-oriented manuscript. A summary of this analysis is provided in the table below.

Table 6: Probability of identifying adult donor and umbilical cord blood match

Population	Description	Any adult donor		Any cord blood			Effective adult donor		Effective Cord blood unit Patient age >=20			Effective Cord blood unit Patient age <20		
		8/8	7/8	6/6	5/6	4/6	8/8	7/8	6/6	5/6	4/6	6/6	5/6	4/6
AAFA	African American	0.33	0.84	0.10	0.76	1.00	0.19	0.71	0.01	0.21	0.77	0.06	0.54	0.94
AFB	African	0.30	0.79	0.08	0.75	1.00	0.16	0.63	0.01	0.20	0.77	0.04	0.53	0.94
CARB	Black Caribbean	0.29	0.76	0.09	0.74	1.00	0.17	0.62	0.01	0.20	0.76	0.05	0.52	0.94
SCAMB	Black South or Central American	0.23	0.61	0.10	0.75	1.00	0.12	0.47	0.01	0.25	0.80	0.06	0.55	0.95
AINDI	South Asian	0.47	0.88	0.19	0.87	1.00	0.33	0.79	0.03	0.35	0.87	0.12	0.69	0.97
FILII	Filipino	0.53	0.86	0.25	0.89	1.00	0.39	0.77	0.03	0.34	0.84	0.14	0.70	0.96
HAWI	Hawaiian/other Pacific Islander	0.35	0.73	0.14	0.79	0.99	0.22	0.60	0.02	0.28	0.81	0.09	0.60	0.95
JAPI	Japanese	0.47	0.84	0.18	0.87	1.00	0.32	0.73	0.03	0.30	0.84	0.11	0.66	0.96
KORI	Korean	0.54	0.90	0.23	0.89	1.00	0.39	0.81	0.04	0.33	0.86	0.14	0.69	0.97
NCHI	Chinese	0.54	0.90	0.25	0.89	1.00	0.39	0.81	0.03	0.34	0.86	0.15	0.70	0.97
SCSEAI	Other Southeast Asian	0.36	0.77	0.16	0.82	1.00	0.25	0.65	0.02	0.31	0.84	0.10	0.63	0.96
VIET	Vietnamese	0.60	0.88	0.25	0.88	1.00	0.45	0.78	0.04	0.33	0.84	0.15	0.68	0.96
MENAF	Mideast/North Coast of Africa	0.56	0.93	0.26	0.90	1.00	0.48	0.89	0.06	0.43	0.90	0.17	0.73	0.98
EURCAU	European Caucasian	0.80	0.98	0.48	0.96	1.00	0.74	0.97	0.14	0.63	0.96	0.34	0.86	0.99
CARHIS	Caribbean Hispanic	0.52	0.88	0.25	0.87	1.00	0.38	0.79	0.05	0.38	0.88	0.16	0.69	0.97
MSWHIS	Mexican or Chicano	0.46	0.88	0.29	0.90	1.00	0.34	0.79	0.06	0.42	0.89	0.19	0.73	0.98
SCAHIS	South or Central Amer. Hispanic	0.41	0.81	0.25	0.87	1.00	0.30	0.71	0.04	0.39	0.88	0.16	0.70	0.97
AISC	Amer. Indian South or Central America	0.47	0.84	0.32	0.89	1.00	0.38	0.77	0.08	0.48	0.91	0.22	0.74	0.98
ALANAM	Alaska Native or Aleut	0.54	0.88	0.24	0.89	1.00	0.38	0.75	0.06	0.42	0.89	0.16	0.72	0.98
AMIND	North American Indian	0.57	0.92	0.33	0.92	1.00	0.47	0.83	0.09	0.50	0.92	0.22	0.77	0.98
CARIBI	Caribbean Indian	0.33	0.72	0.15	0.78	1.00	0.23	0.63	0.03	0.30	0.83	0.10	0.60	0.95

Other analyses that took place under this aim:

- Performed cost-benefit analysis comparing the value of adult donor recruitment versus CBU recruitment. Results indicate that NMDP should have a larger CBU inventory given the current size of the adult donor registry.
- Calculated optimal minimum total nucleated cell (TNC) count for CBU recruitment for adult patients to be 155×10^7 TNC rather than the current HRSA guideline of 90×10^7 TNC.

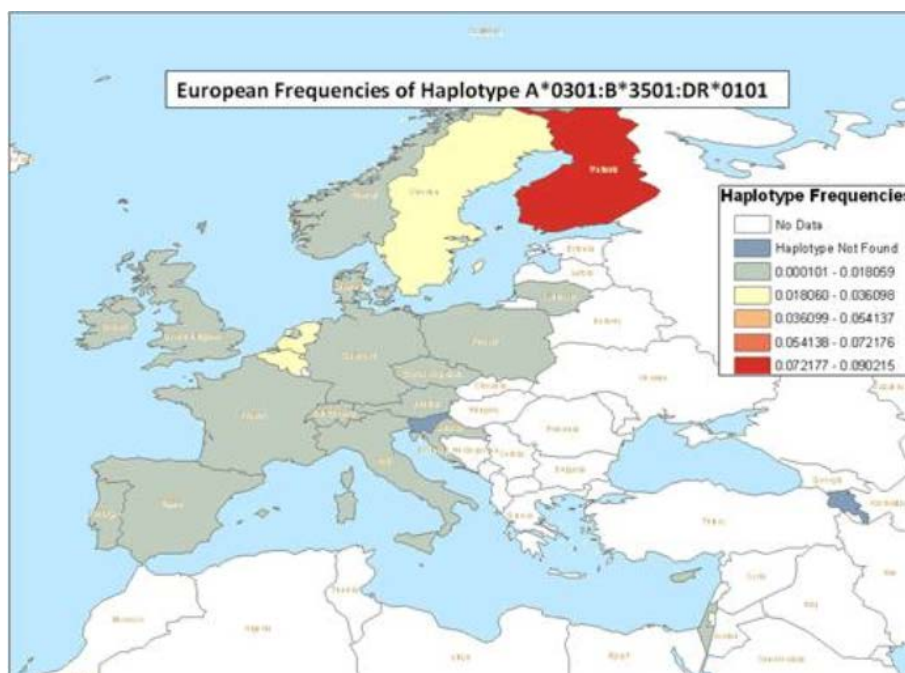
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- Re-calculated HLA match rates for end of year 2010, with several bug fixes implemented. Match rates have been cross-validated with brute-force matching “benchmark” and have been found to be in general agreement at the race category level.
- Calculated historical NMDP match rates back to the beginning of the registry.
- Developed a draft manuscript describing the foundational mathematical model for the registry match rate projection. This manuscript was submitted to the journal Biology of Blood and Marrow Transplant.
- Initiated development of a comprehensive mathematical framework describing HLA high resolution and phase inference for use in different matching models for multiple populations with different self identified race and ethnicities within the Be The Match registry

Aim B.3.4: Target Under-Represented Phenotypes

A new version of the “haplostats” tool (<http://haplostats.org>), which is used by laboratories and investigators worldwide to lookup HLA types and present the corresponding haplotype frequency data in a way that can inform research and medical decision making, was implemented. As an enhancement to this software, we have identified a goal to generate automated maps of HLA haplotype frequencies projected on to maps of the US and the world. We have been working with software tools from ESRI (Environmental Systems Research, Inc.) to automate map production and have completed a prototype of desktop-scale map automation in an easy-to-read world view map.

Figure 27: prototype map of haplotype frequencies in Europe, computed from BMDW



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In an effort to plan future research on ancestry and genetics as it relates to the registry and the need to rapidly identify matched donors, we planned and facilitated a “Genetic Ancestry Summit” in Washington, DC on November 2-3, 2010. The goal of this meeting was to expand the understanding of how ancestry affects HLA. One of the outcomes of this meeting was the development of a research project to:

- Develop a geographical, ancestry-based questionnaire based on origin of parents and grand-parents
- Develop an ancestry information marker (AIM) testing strategy

Aim B.3.5: Bioinformatics Web Site

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.3.6: Maximize software using consultant data

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.3.7: Population Genetics

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.3.8: Haplotype Matching

No funding was requested under this Aim for the 0204 budget cycle.

Aim B.3.9: Global Haplotype/Benchmark

No funding was requested under this Aim for the 0204 budget cycle.

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II.B. Rapid Identification of Matched Donors – Hypothesis 4:

Reducing the time and effort required to identify closely matched donors for patients in urgent need of HSC transplants will improve access to transplantation and patient survival in the context of a contingency response and routine patient care.

Aim B.4.1: Expand Network Communications

Extended the Business Services to support the new alleles and allele combinations expressed as allele codes:

- Limited Support for P codes
- Full support of WMDA approved codes – XXXX, NNNN, UUUU, NEW
- Support in external tools for user queries of allele code information
- Preparation for expansion of allele code information
- Support for new nomenclature vendor DNA typing kits

NMDP collaborated with five Pilot Registries on the European Marrow Donor Information System (EMDIS) cord project by:

- Documenting semantics describing the messages and process flow to exchange inventory
- Documenting fields that each will be sending in the inventory exchange
- Sharing house rules for searching based on CBU status, no differences or concerns

Business-to-Business (B2B) functionality, allowing the exchange of information between organizations was implemented. The B2B components of our inventory exchange model implemented at NMDP allow for the following:

- Ability to support inventory sharing
- Ability to support transaction sharing
- Ability to share NMDP CBU inventory with strategic partners and to keep it updated

Note: Work remains to complete the development of the components required to receive, search and display other Registry cord blood unit inventory.

Aim B.4.2: Central Contingency Management

8/8 High Resolution HLA Match Rate Project:

During this funding period, the NMDP embarked on a critical research project to understand the 8/8 High Resolution HLA Match rate for the four major race/ethnic groups: Caucasian, African American, Asian-Pacific Islander, and Hispanic. Estimation of the 8/8 (A, B, C, DRB1) HR match rate could not be done using real patient unrelated donor (URD) searches, as they present

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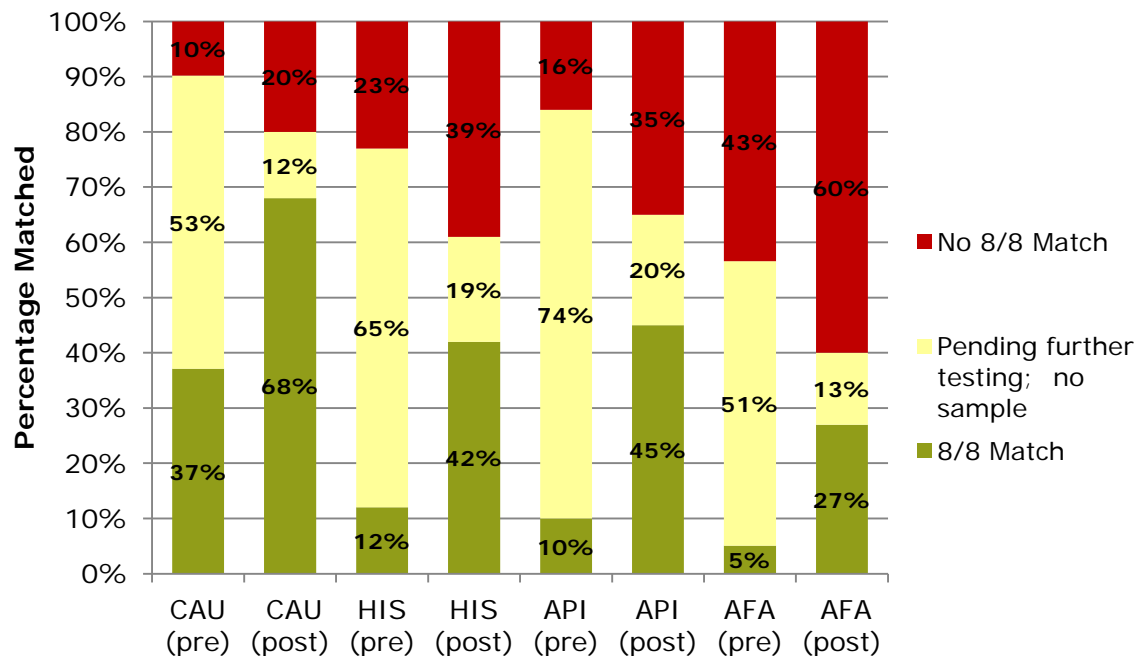
a biased cohort for reasons including access to treatment, financial barriers and incomplete donor testing. This study was designed to estimate the true match rate for the four major race groups, representing historical best and worst match rates, respectively.

URD searches were performed for pseudopatients (PP) who were randomly selected, previously high resolution tested donors in the Be The Match Registry. Searches were based on a fixed registry file as of January 2009. 200 cases that required donor testing were accumulated for each race group. Donors were ranked for likelihood of match and locus specific testing was performed. Donors were tested in rounds for cost efficiency. Donor typing continued until an 8/8 match was identified or no donors remained. Initially only donors with a stored sample were tested.

Approximately 360 NMDP donors had no stored sample for testing. The donor contact team was able to obtain updated addresses for 150 of these donors and they were sent a swab kit with a letter requesting that they return an updated sample for storage and updated HLA typing. 75 donors returned swab kits which were sent out for HLA typing.

The final 8/8 Match Rate results for each race group, pre and post- HLA typing are displayed in Figure 28. CAU have the highest match rate at 68%, followed by API at 45%, HIS at 42%, and AFA at 27%. These results provide valuable information for physicians' education of patients in their likelihood of identifying an 8/8 match.

Figure 28: 8/8 Donor Match Rate by Race Group



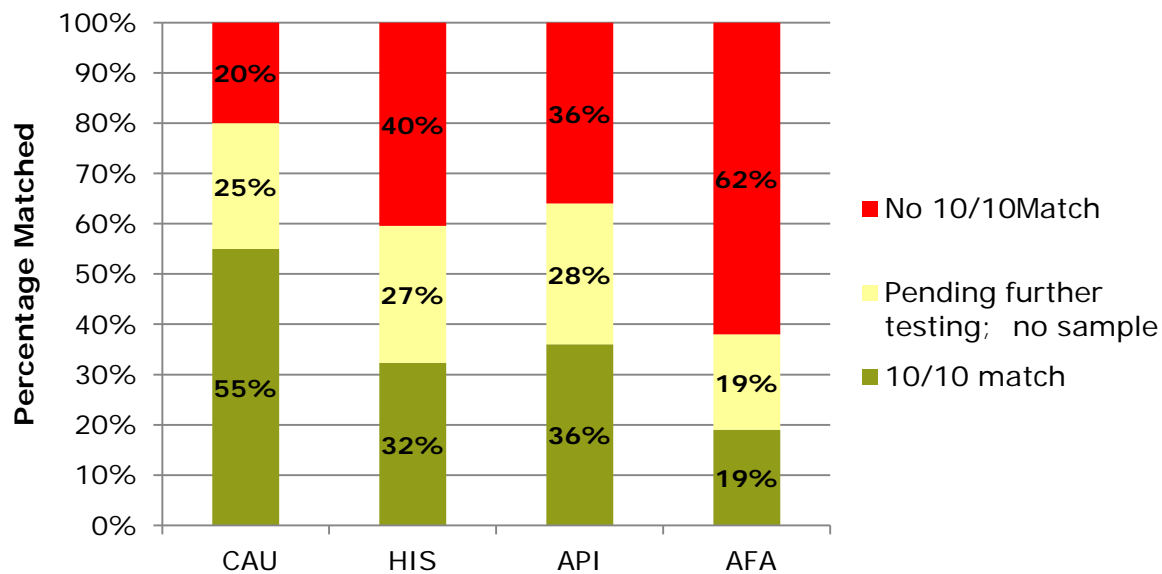
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Development of a manuscript for this study is in progress with plans to submit to a peer-reviewed journal. In addition, an abstract was accepted as an oral presentation at the ASBMT Tandem meeting in February 2011.⁷

A poster abstract was also presented at the 2011 EFI meeting in May 2011, which included project data on the efficiency of 8/8 donor selection.⁸

During the course of presenting data, centers inquired about the 10/10 donor match rates, as they often use 10 allele matching when selecting donors for a patient. The 8/8 study was expanded to analyze 10/10 high resolution matches (adding DQB1) on patients where an 8/8 match was identified. The results are shown in Figure 29, although depletion of donor samples prevented the full resolution of DQB1 typing for many donors.

Figure 29: 10/10 Donor Match Rate by Race



Aim B.4.3 Conduct a transplant center benchmarking analysis to identify center-specific factors (e.g., quality management techniques and processes) that contribute meaningfully to superior survival outcomes. Share processes that contribute to superior outcomes with the entire TC network as best practices.

No funding was requested under this Aim for the 0204 budget cycle.

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Aim B.4.4 Identify plans to expand capabilities of collection center and apheresis center network to meet increasing number of donor product requests on both a short-term and long-term basis.

No funding was requested under this Aim for the 0204 budget cycle.

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II.C. Immunogenetic Studies – Hypothesis 1:

HLA mismatches may differ in their impact on transplant outcome, therefore, it is important to identify and quantify the influence of specific HLA mismatches. In contingency situations it will not be possible to delay transplant until a perfectly matched donor can be found.

Aim C.1.1: Donor Recipient Pair Project

A retrospective Donor/Recipient (D/R) Pair HLA typing project to perform high resolution class I (HLA-A, B, and C) and class II (HLA-DRB and DQB1) typing of paired samples from NMDP's Repository, was initiated in 1994. The primary objectives of the D/R Pair Project are to:

- Determine the impact of DNA-based HLA matching on unrelated donor transplant outcome
- Develop strategies for optimal HLA matching
- Evaluate the impact of matching at alternative HLA loci on transplant outcome
- Promote the development of DNA-based high resolution HLA typing methodologies

Transplant pairs were chosen from stored samples at the NMDP Research Sample Repository and distributed to participating laboratories for high resolution HLA typing. All paired samples are selected in collaboration with the CIBMTR Statistical Center to ensure the additional cases would benefit ongoing and future analyses. The cohorts tested during the project period consisted mainly of transplants that utilized peripheral blood stem cells as the cell source, reduced intensity or non-myeloablative preparative regimens, rare diseases and older patients, reflecting the expanding indications for unrelated donor Hematopoietic Stem Cell Transplant (HSCT). In addition, the project has added CBT pair samples to facilitate studies of HLA matching in this high growth field.

Testing was completed on an additional 146 donor/recipient and 192 cord/recipient pairs during the project period, bringing the total enrolled to over 15,000. Typing results were reported electronically to the NMDP and compared with previous transplant center results as a measure of quality control. At the initiation of the project period, four laboratories participated in the project. Following a competitive bid processes, the laboratory network remained the same, with four laboratories and pricing that was level with the last bidding cycle. Typing at the DPB1 locus was added back due to the increased interest in studies with DPB1 typing. Presence/absence KIR genotyping on 2DL1-5, 2DS1-5, 3DL1-3 and 3DS1 with has continued. To date, over 2100 pairs and 1180 additional donors have been typed for presence/absence of 14 KIR loci (2DL1-5, 2DS1-5, 3DL1-3 and 3DS1).

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In order to continuously upgrade the D/R Pairs Project, and include as many cord/recipient pairs we have begun to include IDs who have Whole Genome Amplified (WGA) DNA as a sample choice. Samples of recipients and cords with limited blood or DNA samples in the research repository were selected and sent for WGA. WGA of these samples allowed for the inclusion of 98 cord/recipient pairs during the project period.

The high resolution HLA data generated through the project are routinely incorporated into all outcomes analyses performed by the NMDP/CIBMTR to provide the best HLA typing and matching information possible. The project has developed the largest fully validated pool of unrelated stem cell transplant donor-recipient HLA data in the world and is an unparalleled resource for transplant research. The data generated through the project have had a major impact on the evolution of the NMDP HLA matching requirements.

Current HLA matching guidelines for unrelated HCT recommend avoidance of mismatches only within the antigen recognition site, i.e. exons 2 and 3 for HLA class I and exon 2 for HLA class II. This recommendation is based on the hypothesis that amino acid differences outside the antigen recognition site are not immunogenic. There is little functional data available to prove this hypothesis and clinical analysis would require an unattainable data set to reach significance, as previously reported.⁹ In brief, an investigation of DRB1*140101 and *1454 mismatches was performed. From a pool of 4222 8/8 matched European American donor/recipient transplant pairs in the NMDP database, only 102 pairs were identified that carried the unresolved DRB1*1401/DRB1*1454 with matching at class I loci. The DRB3 linkage was used to identify 12 pairs likely to be mismatched for DRB1*140101/DRB1*1454, but was determined an insufficient sample size to assess the impact of the mismatch on transplant outcome.

The Antigen Recognition Site Allo-reactivity Assessment Project will give insight into the allowable tolerance of matching needed outside of this binding region. Specific queries of the Be The Match Registry allowed for selection of ninety-nine potential donors from 4 of the 12 haplotypes identified to be typed at high resolution. Typing of the HLA-A, B, C, DRB1/3/4/5, DQA/B1, and DPB1 genes was completed on all 99 donors. During this period, potential donors representing the 4 different 7/8 mismatch haplotypes were invited to participate in the Antigen Recognition Site Allo-reactivity Assessment Project. 72 donors of the 99 were invited to participate in the study. 21 study participants consented and submitted blood samples. Samples were drawn, processed, and shipped for inclusion in in-vitro functional cellular assays. Eleven samples of four different haplotype pairs were shipped to be tested. The testing was completed during the period of performance from May 9, 2011 to August 31, 2011. Data analysis is underway and will be completed under subsequent funding cycles.

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II.C. Immunogenetic Studies – Hypothesis 2:

Even when patient and donor are HLA matched, GVHD occurs so other loci may play a role.

Aim C.2.1: Analysis of non-HLA loci

Recent research has heightened interest in additional genetic polymorphisms which may modify the outcomes of transplantation. HLA genes other than the major histocompatibility complex (MHC) found on chromosome 6 and non-HLA genetic factors may all influence the suitability and success of allogeneic stem cell transplants. The largest body of data with clear correlation to unrelated stem cell transplant outcome was surrounding the role of Natural Killer (NK) cells. These cells express inhibitory receptors (KIR) that specifically interact with MHC class I molecules. Genes encoding for these Ig-like ligands are found on chromosome 19. The regulatory mechanism mediated by these receptors is thought to protect normal cells from autologous NK attack, while rendering cells for which class I expression is compromised (e.g. by tumor transformation or viral infection) or incompatible (e.g. by stem cell transplant) susceptible to NK-mediated killing. This has been shown to be responsible for anti-leukemic effects and protection against GVHD following allogeneic HSC transplantation.

Based on this information, the NMDP developed a pilot study to perform KIR typing utilizing selected donor and recipient pair samples. The project was launched in early 2005 with ongoing support provided through the project period. The NMDP selected three laboratories to participate in the project through a competitive bid process. The primary objectives of the study were to:

- Move technology forward from the current practice of locus level typing to high resolution typing
- Disseminate information and protocols in an open source mechanism
- Develop reference lines for use in individual laboratories. Additionally, the project will provide more fully characterized and highly quality controlled transplant pairs for use in research studies connecting these factors to clinical outcome data.

Previously, the KIR Typing Pilot Project completed the final typing on 435 Caucasian donor samples for 14 KIR genes (2DL1-5, 2DS1-5, 3DL1-3 and 3DS1). During this period of performance, final characterization of 46 new alleles occurred. Publication of the new Immuno-Polymorphism Database (IPD) containing these alleles is expected within the next year. Presence/absence genotyping of the KIR loci in the retrospective D/R Pair HLA typing project (II.C.1) has continued. To date, over 2100 pairs and 1180 additional donors have been typed for presence/absence of 14 KIR loci (2DL1-5, 2DS1-5, 3DL1-3 and 3DS1).

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The results of the KIR Typing Pilot Project were presented as three separate abstracts at two different meetings. “Refinement of KIR high resolution haplotypes and patterns of linkage disequilibrium in Caucasians” and “A web application for calculating KIR haplotype frequencies” were presented at the 2011 KIR Polymorphism Workshop in Tammsvik, Sweden. “A web application for calculating allelic killer immunoglobulin-like receptor (KIR) haplotype frequencies and linkage disequilibrium statistics” was presented at the 2011 ASHG Meeting in Montreal, Canada. The availability of reference cell lines and data were promoted to external researchers during both meetings. The clinical correlation of KIR alleles with HCT outcome was initiated in collaboration with the International Histocompatibility Working Group; HCT component and analyses are ongoing.

NK cells have also been implicated in unrelated HSCT outcome through suppression of GVHD, promotion of HSC engraftment, and mediation of graft versus leukemia effects. NK-HLA interaction through inhibitory KIR has been a major focus of investigations regarding the role of NK in HSCT.

The Immunobiology Project Results (IPR) database and its applications allow for storage and analysis of all immunogenetic data collected on NMDP research samples. This database replaces the previous HLA D/R pairs database and facilitates storage and analysis of data from other immunogenetic loci (KIR, microsatellites, single nucleotide polymorphisms, etc).

We have implemented this software and development a number of minor releases that include new functions and enhancements.

We have expanded the Immunobiology Integrated Database (IIDB) to include the population of data for the subject areas Match Grades, Match Grade Variables and Infectious Disease Markers and new ID fields were added to facilitate the linking of NMDP and CIBMTR data. The design of this database was refactored to make it compliant with NCI Biomedical Research Integrated Domain Group (BRIDG) standards.

Building upon the loading and validation of the typings, this period’s work emphasized the migration of historical data and the implementation of graphical web tools and reports that allow business users to resolve discrepancies between typings and set the audit flags. The transition to database 3.1.0 of the HLA nomenclature was completed. The ‘look and feel’ of the application was improved, and the contents of allele codes can now be viewed by hovering the mouse over them.

An ID report was created to allow users to determine what SG certain IDs are incorporated in. Finally many bug fixes were addressed.

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Additional accomplishments in this aim included:

- Naming, submission and publication of 46 novel alleles in 78 samples from KIR Typing Pilot.
- Distribution of a second version of the manuscript for the KIR Typing Pilot Project to external co-authors. Further analysis of the data is ongoing. Presentation of the data will occur at the end of next quarter.
- Typing of 2400 pairs and 1200 donors from the D/R pair's project for presence/absence of 14 KIR loci (2DL1-5, 2DS1-5, 3DL1-3 and 3DS1).

Immunobiological test results generated through NMDP/CIBMTR approved studies and reported to the NMDP are summarized in Table 7. These data will be used for testing, validation, and population of the IPR database.

Table 7. Immunobiology typing projects utilizing NMDP samples and contributing data to the IPR database

Study Title	Investigator	Number of Samples	Genes of interest	Testing Method	Data Submitted
NK Cells, Their Receptors and Unrelated Donor Transplant ^{10,11}	J. Miller	2300 pairs	KIR	RT-PCR, FACS, SSO, MALDI-TOF	Yes
Survey of Diversity of Immune Response Genes in Unrelated Hematopoietic Stem Cell Transplantation	C. Hurley	40 Pairs	cytokine and KIR	SBT	Yes
Candidate Gene Study to Examine the Impact of Chemokine and Chemokine Receptor Gene Polymorphisms on the Incidence and Severity of Acute and Chronic GVHD ¹²	R. Abdi	1300 pairs	CCL1, CCL2, CCR5, CCR2, CX3CR1	Taqman PCR	Yes
Functional Significance of Killer Ig-like Receptor (KIR) Genes in HLA Matched and Mismatched Unrelated HCT ¹³	B. Dupont, K. Hsu	2000 pairs	KIR	SSP	Yes
Functional Significance of Cytokine Gene Polymorphism in Modulation Risk of Post-Transplant Complications	E. Petersdorf	2500 pairs	>30 Immune response genes	Taqman PCR	Yes

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Study Title	Investigator	Number of Samples	Genes of interest	Testing Method	Data Submitted
Identification of Functional SNPs in Unrelated HCT ^{14,15}	E. Petersdorf	3500 pairs	Entire MHC region	Taqman PCR	In Process
Use of Female Donors with Pre-existing Antibody to H-Y Antigen will Result in Robust Serologic Response to H-Y Antigens in Male HSC transplantation Recipients	D. Miklos	288 pairs	H-Y Antigen	ELISA, protein array	Yes
Multiplexed Genotyping of Human Minor Histocompatibility Antigens (mHAg): Clinical Relevance of mHAg Disparity in Stem Cell Transplantation ¹⁶	T. Ellis	730 pairs	mHAg	Allele-specific Primer Extension	Yes
Genetic Polymorphisms in the Genes Encoding Human Interleukin-7 Receptor- α : Prognostic significance in Allogeneic Stem Cell Transplantation ¹⁷	K. Muller	851 pairs	IL-7	Taqman PCR	Yes
The Effect of Non-Inherited Maternal Antigens in Cord Blood Transplantation ¹⁸	L. Baxter-Lowe	102 pairs	HLA	SBT	Yes
Detection of HLA Antibody in Single Antigen HLA-Mismatched Unrelated Donor Transplants	S. Arai, D. Miklos	200 pairs	Anti-body	ELISA, Protein array	Yes
Detection of Donor-Directed, HLA-Specific Alloantibodies in Recipients of Unrelated Stem Cell Transplantation and Their Relationship to Graft/Patient Outcome ¹⁹	R. Bray	111 pairs	Anti-bodies	Flow cytometry	Yes
Genome-wide Association in Unrelated Donor Transplant Recipients and Donors: A Pilot Study	R. Goyal	858 pairs	> 600,000 Genome wide SNPs	Human 610 - Quad V1 arrays	In process
SNPs in the p53 Pathway and Outcomes in URD HCT	B. DuPont	1500 pairs	p53, ATM, MDM2 and p21/Waf1	Taqman	In process

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Study Title	Investigator	Number of Samples	Genes of interest	Testing Method	Data Submitted
Association of Donor and Recipient Gene Polymorphisms of Drug and Innate Immune Response with Outcomes after URD HCT	V. Rocha	725 pairs	GSTP, GSTT, GSTM, UGT CD14, TIRAP, and NALPs	Taqman	In process
To Develop and Test a Prognostic Index for Survival in CML URD HCT ²⁰	A. Dickinson	1100 pairs	TNF, IL-1RA and IL-10	Taqman	Yes
Evaluation of TGF-β1 Promoter and Signal Peptide Polymorphisms as Risk Factors for Renal Dysfunction in HCT Patients Treated with Cyclosporine A ²¹	R. Shah	400 samples	TGF-β1	Taqman	Yes
Donor and Recipient Telomere Length as Predictors of Outcomes after Hematopoietic Stem Cell Transplant in Patients with Acquired Severe Aplastic Anemia	S. Gadalla	650 samples	Telomere length and Telomerase Polymorphisms	Taqman	In process
Development of a GVHD Prevention Biodiagnostic Test	R. Somogyi	450 samples	Gene Expression Array	Array	In process
Genetic polymorphisms and HCT related mortality Re: Pre-HCT conditioning in matched unrelated donor HCT	T. Hahn	>4,000 pairs	GWAS	Array	In process
Impact of CTLA4 SNPs on outcome after URD transplant	M. Jagasia	1,200 pairs	CTLA-4 SNPs	Taqman	In process
KIR genotyping and immune function in MDS patients prior to unrelated donor transplantation	A. E. Warlick and J. Miller	970 samples	KIR genotype, expression and cellular function	SSP, flow cytometry and cellular assays	In process
Plasma YKL-40 and CHI3LI genotype to predict mortality after unrelated donor HCT	B. Kornblit	800 pairs	YKL-40 plasma levels and CHI3LI SNPs	ELISA and Taqman	In process

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Study Title	Investigator	Number of Samples	Genes of interest	Testing Method	Data Submitted
Natural killer cell genomics and outcomes after allogeneic transplantation for lymphoma	V. Bachanova, J. Miller, D. Weisdorf and L. Burns	800 pairs	KIR genotype, expression and cellular function	SSP, flow cytometry and cellular assays	In process
Effect of genetic ancestry matching on HSCT outcomes	A Madbouly, M. Maier and N. Majhail	2300 pairs	Ancestry Informative Markers	Taqman	In process

Aim C.2.2: Related Pairs Research Repository

No funding was requested under this aim for the 0204 budget cycle.

Aim C.2.3 CIBMTR Integration

No funding was requested under this aim for the 0204 budget cycle.

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II.D. Clinical Research in Transplantation – Hypothesis 1:

Clinical research in transplantation improves transplant outcomes and supports preparedness for a contingency response.

Aim D.1.1: Observational Research, Clinical Trials, and NIH Transplant Center

Resource for Clinical Investigations in Blood and Marrow Transplantation

During this grant period, the RCI BMT continued to work towards its goal to provide an avenue for investigators to obtain statistical and data management support for prospective trials and projects in HCT. The following key elements were completed:

- Clinical Trials Advisory Committee (CTAC) held two annual in-person meetings during this grant period. Both meetings occurred during the Tandem meetings, in February 2011 and 2012. This committee has been charged with providing scientific review and recommendations on clinical trial proposals. The committee reviewed two proposals and one was approved to move forward to protocol development. The CTAC recommended the second proposal not proceed because of concerns that accrual could not be achieved in a reasonable time period.
- Accrual was completed on the Adult Double Cord protocol for patients with hematologic malignancies on September 13, 2011. A total of twelve sites enrolled 56 patients during the course of this trial of which the last 20 occurred during this grant period. Staff continued working with sites to ensure all data was submitted, data queries were addressed and performed site monitoring.
- Staff continued to provide support to the Blood and Marrow Transplant Clinical Trials Network (BMT CTN) PBSC vs. Marrow Phase III trial. This support included managing the donor component of the study. Staff performed monitoring activities at the donor centers. During this grant period a total of 20 monitoring visits occurred with 320 of the 551 donor data sets were monitored.
- The Long-Term Donor Follow up (LTDFU) study opened October 1, 2010. During this grant the logistics and processes were established. Accrual to this study included donors who previously donated in addition to donors who are donating currently. As of the end of this grant period a total of 10,838 donors had consented to participate. This is 34% of the goal of 32,128 donors. The Survey Research Group (SRG) staff is responsible for the follow up contacts for the NMDP operated donor centers. During this grant period SRG completed 3,428 follow up calls.

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Cord Blood Research Activity

The subcommittee membership continued work on several ongoing research projects. A study team developed a study entitled “Cord Blood Biomarkers for Engraftment,” which focused on the evaluation of the enzyme aldehyde dehydrogenase (ALDH) expression as a reliable marker for stem cell potency. The primary hypothesis was that the ALDH bright (ALDHbr) dose, measured in a frozen segment from a banked CBU, would best correlate with engraftment after transplantation. If validated, it would provide a method to assay CBU potency before release of a unit to a transplant center. It would also result in selection of higher quality CBUs for CBT. Work continued with project funds to validate the testing methodologies at the two centralized laboratories, Carolinas Cord Blood Bank at Duke and MD Anderson Cancer Center (MDACC) to ensure consistent inter-laboratory results; however, initial statistical analysis of the validation testing results showed poor inter-laboratory reliability for all assays performed (Table 8).

Table 8: Inter-laboratory reliability measurements for 104 assayed segments

	Reliability	
Measurement	Raw values	Log transformed values
Total gated cells	0%	0%
Total ALDHbr	39%	67%
Total CD45	0%	0%
Total CD34	0%	0%
Viable CD45	0%	0%
ALDHbr (Viable CD45)	38%	65%
ALDHbr (% viable CD45)	75%	73%
Viable CD34	7%	15%
ALDHbr (Viable CD34)	11%	50%
ALDHbr (% viable CD34)	31%	54%
ALDHbr (Viable CD34 and CD45)	0%	0%

As a result of the poor inter-laboratory reliability, the study team held multiple conference calls to discuss the possible areas in which variation could have been introduced into the protocols of the assays. Further protocol development and testing was then completed at Duke and showed improved intra-laboratory reliability for the ALDHbr assay. The study team next planned for a validation assessment between Duke and MDACC using the revised protocol to determine study

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progression feasibility. Upon completion of the additional testing, poor inter-laboratory reliability was found to be persistent (Table 9).

Table 9: Inter-laboratory reliability measurements for 45 assayed segments.

Source	ALDHBR (% of viable CD45)	ALDHBR (% of viable CD34)	ALDHBR (% viable CD34 and CD45)
Cord	0.06098	132.2068	0
Error	0.03378	306.9035	389.91075
Reliability	64%	30%	0%

Due to persistent reliability issues between the testing sites of Duke and MDACC during the validation phase of the study, a third testing site, St. Louis Cord Blood Bank (SLCBB), was introduced to determine the feasibility of attaining inter-laboratory results correlation. Contract negotiations with SLCBB were initiated and finalized. The study group determined the training and validation plans and leased the appropriate flow cytometer for the SLCCB laboratory to perform the assays within the specifications of the manufacturer.

Work continued on an observational study of single versus double CBT in adults. Further analyses to expand the cohort to include participants from 2007 and 2008 were requested and completed. The principal investigator, EJ Shpall, MD, presented the results at the NMDP Cord Blood Advisory Group meetings in both June and October of 2010. A draft manuscript is in process.

A retrospective study to evaluate the benefit of matching for non-inherited maternal antigens (NIMA) in CBT was developed and completed during the project period. An example of NIMA matching at HLA-A is depicted in Table 10 below. The recipient is mismatched with the cord blood unit at HLA-A: A*01 versus A*68. The recipient does have NIMA of A*01. This was considered a NIMA match.

Table 10. Example of NIMA matching at HLA-A

Typing	HLA-A	HLA-A	HLA-B	HLA-B	HLA-DRB1	HLA-DRB1
Maternal	01	68	08	58	0301	1201
Recipient	01	74	45	58	1201	1201
Cord Blood	68	74	58	58	1201	1201

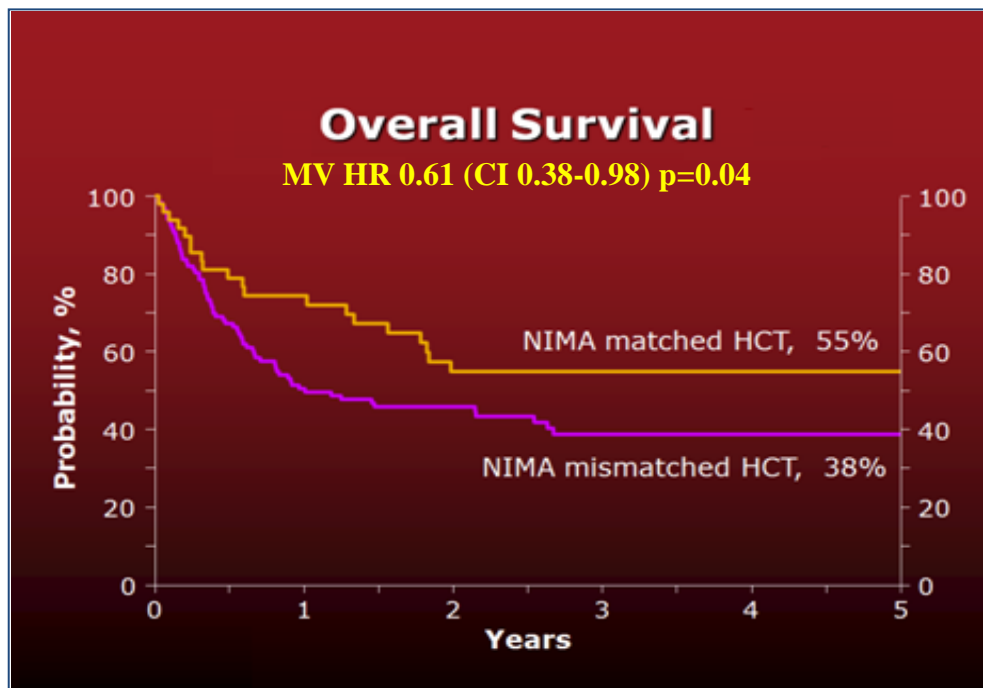
Maternal samples and HLA typing data were collected from participating cord blood banks (CBB) and HLA typing performed to determine the NIMA match status of the cases enrolled in

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the study. Collaboration was initiated with Eurocord to increase the size of the cohort with each group contributing approximately 250 cases. The HLA typing data from CIBMTR and Eurocord was collated and coded for HLA and NIMA match grades for each case in the cohort. The HLA data was merged with the clinical outcome data and analyzed. An abstract was presented at the 2011 EBMT annual meeting.²²

The final analysis included 508 patients with hematologic malignancies who received a 5/6 or 4/6 CBT were included in the initial dataset and 52 were NIMA matched (10%). A matched pair analysis was conducted with 48 NIMA matched patients (NIMA+) matched to 116 NIMA mismatched patients (NIMA-) at a ratio of 1, 2 or 3 controls per case. Cases and controls were matched for disease, disease status, HLA match, patient age, cell dose, and conditioning regimen. Four NIMA matched cases could not be matched to controls and were not included in the final analysis. No differences were found in neutrophil recovery and acute and chronic GVHD between the two groups; however, transplant related mortality was significantly lower in NIMA+ patients ($p=0.05$) versus NIMA- leading to a superior 5 year probability of overall survival (55% NIMA+ vs. 38% NIMA-) (Figure 30). These data suggest that when faced with the choice between multiple HLA-mismatched UCB units containing sufficient cell doses, selecting a NIMA+ UCB unit may improve survival after mismatched UCB transplant.

Figure 30. The 5-year probabilities of overall survival after NIMA matched and mismatched transplantation.

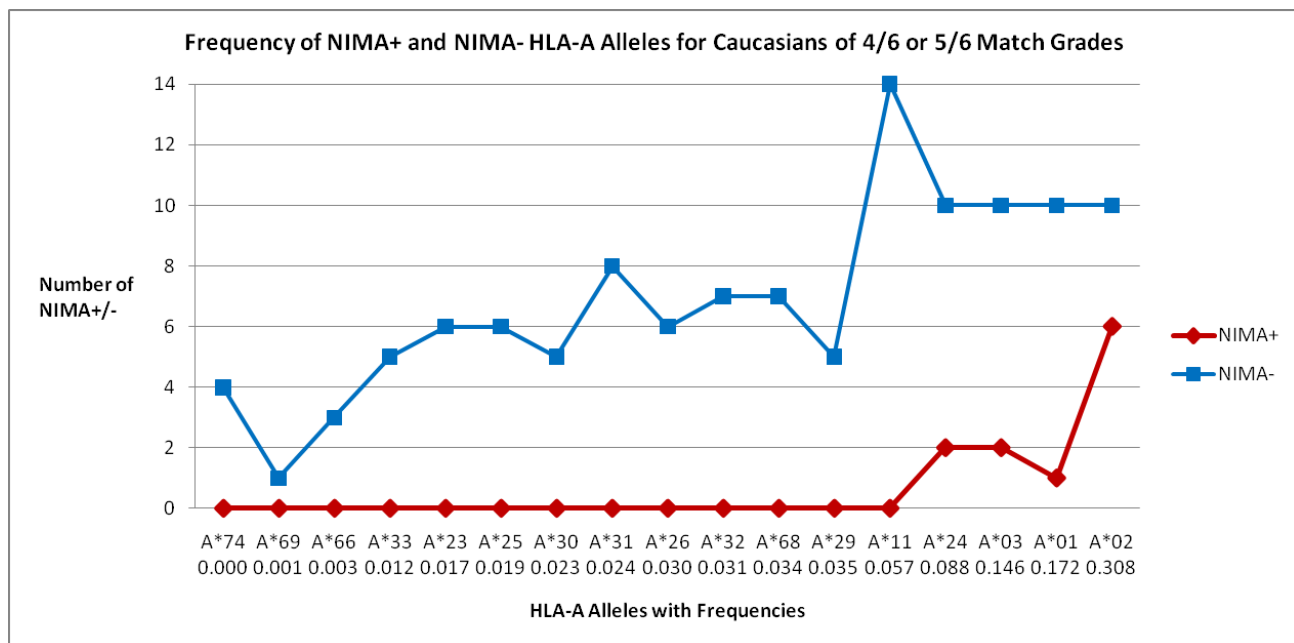


In addition to the clinical outcomes evaluation described above, an analysis was conducted on the likelihood of identifying NIMA matches based on the HLA genotype of the patient. The results were presented in an abstract entitled: "The Influence of HLA Antigen/Allele Frequency

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on Access to Non-inherited Maternal Antigens for Recipients of CBT Mismatched at HLA-A, B Antigen or DRB1 Allele Levels” at the 9th annual International Umbilical Cord Blood Transplantation Symposium in 2011.²³ The study was chosen as the Best Abstract of the Symposium by the Abstract Review Committee and awarded an oral presentation during a general session. The main conclusion of the analysis was that NIMA matches are unlikely to be found for low frequency HLA alleles and search strategies should focus on mismatching for higher frequency alleles to maximize the potential to identify a NIMA match (Figure 31).

Figure 31. Distribution of NIMA matched and mismatched frequencies for Caucasian CBT pairs at HLA-A



Work was completed on a white paper detailing recommendations/guidelines for the assessment of new assays (potency or other assays) relevant to cord blood banking and/or transplantation. A white paper was published in Cytotherapy¹ and focused on the following topics:

- A brief history of CBT, with a focus on cord blood characteristics (ex. TNC) and recipient clinical indicators (ex. Degree of HLA match, diagnosis) that impact outcomes
- A review of the key steps in CBU manufacturing (collection, processing, cryopreservation, storage) and handling (ex. Transportation, short-term storage, thawing) that can impact unit potency
- A description of assays currently used to assess CBU quality
- Expectations for the next-generation of assays, including how the assays should be validated, implemented and any regulatory implications

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The laboratory staff from the cord blood banks of Duke, MDACC, Puget Sound, and SLCBB began working with Stem Cell Technologies to assess the efficacy of STEMvision, an automated CFU enumeration instrument. Work continued and was finalized during the next project period.

Work began on the pilot study, “Exchange, analysis and standardization of cord blood CD34+ cell counts using ImmPort Flow Cytometry Analysis Component (FLOCK),” designed to assess a system for centralized flow cytometry based CD34 analysis. Flow cytometry data files from the 2009 and 2010 proficiency testing samples of multiple laboratories were collated and an attempt was made to analyze them using FLOCK. St. Louis and Puget Sound laboratory staff worked with the developers at ImmPort on flow file upload for inter- and intra-bank data analysis. Differences in flow instrumentation and software between the two banks proved problematic and ultimately put the project on hold until further optimization of the FLOCK software can support these differences.

NIH Search Support

The National Institutes of Health (NIH) has been accepted as an NMDP transplant center since 2007. Prior to that time, the NIH, representing our Nation’s premier medical research endeavor, was not applying their considerable problem-solving skills to issues surrounding unrelated donor transplantation. The NMDP, with ONR support, set out to remedy that deficiency by entering into collaboration with NIH. This collaboration has been extremely successful.

The NMDP is collaborating with intramural NIH transplant programs from the NCI, the NHLBI and the NIAID. These programs are investigating alternative approaches in unrelated donor transplantation to improve patient outcomes. The actual transplants and the investigational portions of each transplant (i.e., the research protocols) are supported entirely with NIH funds. Navy funding supplies support for donor identification, selection, and collection. NMDP donors are not research subjects on these protocols because the donors are making standard donations for accepted transplant indications. The research component of these transplants is conducted entirely by NIH intramural program staff and funded entirely with NIH dollars. The NMDP provided support for the collection of 40 products (26 PBSC, 1 bone marrow, 11 CBU, and 2 therapeutic T cell) under the grant.

CIBMTR Observational Research

Support of the Observational Research program included statistical hours for managing studies within the Immunobiology (see section IID1.3 below), GVHD, and Graft Sources Working Committees. During this grant period staff performed proposal review, protocol development, data preparation, data analysis, and manuscript preparations. Details regarding the Immunobiology activities can be found in IID1.3 below. The GVHD and Graft Sources Working Committees published 11 manuscripts.²⁴⁻³⁴ During the grant period, staff performed various other functions on over 20 other studies.

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FormsNet Development

- Four successful production releases were completed supporting the reporting and business requirements for the LTDFU and MDS trials.
- Migration of Legacy NMDP Recipient forms into the FormsNet 2 database has progressed. A testing approach was developed and proved. Conversion executed in January 2012.
- Business approval and tech review of the Business Requirements Definition (BRDs), Design and Implementations were completed for:
 - Killer Immunoglobulin-like Receptor Donor Selection (KIR-DS), CIBMTR Sample Tracking Application, 09 Minimal Residual Disease (MRD) Study
 - Sample Tracking Application to support MRD Study
- All Audit Recipient module development milestones were completed and implemented.
- Milestones for FormsNet3 planning were created. The following were completed:
 - Forms Engine Part 1 BRD and Validation BRD review
 - Form Specification template format definition completed
 - FN3 charter approved
 - Donor Audit BRD approved
 - Scanning/Imaging tool BRD approved
 - FN3 application design

A Growable Network Information System (AGNIS)® Development

- Authorized five additional transplant centers to retrieve form data using the Stemsoft BMTBase 4.0 product, worked with 3 centers that had previously been set up to correct issues with their access.
 - A total of 18 centers were authorized for form retrieval
 - 15 of these centers have retrieved completed forms through this AGNIS interface.
- Completed development, and quality assurance of the 100 day Post-HSCT comprehensive Follow-up form. This form was implemented.
- Improved error handling within the AGNIS publish function, (issues encountered had prevented forms from being published to the AGNIS repository).
- Provided support for Memorial Sloan Kettering on their AGNIS development efforts, they successfully submitted forms to the AGNIS external development environment.

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- Provided support to EBMT development and form mapping efforts. EBMT completed their mapping to the Pre-Transplant Essential Data form. They have begun testing of their Post-Transplant form mappings (completed and submitted into production in 2012).
- Implemented coding of AGNIS changes to accommodate assignment of patient unique identifier
- Implemented coding of AGNIS changes to allow submission and retrieval of forms prior to form curation in the Cancer Data Standards Repository (caDSR); quality assurance of this feature is still outstanding

Aim D.1.2: Research with NMDP Donors

No funding was requested under this aim for the 0204 budget cycle.

Aim D.1.3: Expand Immunobiology Research

During a previous grant period, the NMDP developed the Immunobiology Research grant request and award procedures for use by the IBWC and developed the IBWC Web site (<http://www.cibmtr.org/Studies/Immunobiology/Pages/index.aspx>). The content was further refined and migrated to the new CIBMTR.org Web site (<http://www.cibmtr.org>) during the grant period.

Grant funds supported significant outreach efforts by the IBWC leadership to increase exposure for the IBWC to basic scientists. The IBWC leadership revised and reprinted the IBWC brochure and informational materials for distribution at scientific meetings and had a presence at the American Society of Hematology, BMT Tandem, European Group for Blood and Marrow Transplant, International Histocompatibility and Immunogenetics Workshop, European Federation of Immunogenetics and Cord Blood Symposium meetings. Support permitted the committee to maintain a strong performance record with 10 abstracts, 9 publications (submitted or accepted) and collaboration on 3 grants completed in calendar year 2011. In addition, 6 new proposals were accepted by the IBWC during the BMT Tandem meetings in February 2012.

IBWC 2011 manuscripts (submitted/accepted):

1. Valcárcel D, Sierra J, Wang T, et al. One antigen mismatched related vs. HLA matched unrelated donor hematopoietic transplantation in adults with acute leukemia: CIBMTR results in the era of molecular typing. *Biol Blood Marrow Transplant*. 2011;17(5):640-648.
2. Woolfrey A, Klein JP, Haagenson M, et al. HLA-C Antigen mismatches are associated with worse outcomes in unrelated donor peripheral blood stem cell transplantation. *Biol Blood Marrow Transplant*. 2011;17(6):885-892.

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3. Dong L, Wu T, Gao ZY, et al. The outcomes of family haploidentical hematopoietic stem cell transplantation in haematological malignancies are not associated with patient age. *Biol Blood Marrow Transplant*. 2011;17(8):1205-1213.
4. Marino SR, Lin S, Maiers M, et al. Identification by random forest method of HLA class I amino acid substitutions associated with lower survival at day 100 in unrelated donor hematopoietic cell transplantation. *Bone Marrow Transplant*. 2012;47(2):217-26.
5. Spellman S, Klein JP, Haagenson M, et al. Scoring HLA Class I Mismatches by HistoCheck Does Not Predict Clinical Outcome in Unrelated Hematopoietic Stem Cell Transplantation. 2012;18(5):739-46. Epub 2011 Sep 29.
6. Rocha V, Spellman S, Zhang MJ, et al. Effect of HLA-matching recipients to donor non-inherited maternal antigens on outcomes after mismatched umbilical cord blood transplantation for hematologic malignancies. Submitted.
7. Pearce KF, Lee SJ, Haagenson M, et al. Analysis of non-HLA genomic risk factors in HLA-matched unrelated donor hematopoietic cell transplantation for chronic myeloid leukemia. *Hematologica*. Epub 2012 Jan 22.
8. Fleischhauer K, Shaw B, Gooley T, et al, on behalf of the International Histocompatibility Working Group in Hematopoietic Cell Transplantation. Effect of T-cell-epitope matching at HLA-DPB1 in recipients of unrelated-donor haemopoietic-cell transplantation: a retrospective study. *Lancet Oncol*. 2012;13(4):366-74. Epub 2012 Feb 15. Submitted 2011.
9. Venstrom JM, Pittari G, Gooley TA, Chewning J, Spellman S, Haagenson M, Gallagher MM, Malkki M, Petersdorf E, Dupont B, Hsu KC. Donor activating KIR2DS1 protects against acute myeloid leukemia relapse in an HLA-dependent manner. Submitted.

IBWC 2011 Abstract presentations:

1. Woolfrey A, Horan J, Wang T, et al. Evaluation of HLA matching requirements in unrelated hematopoietic stem cell transplantation for non-malignant disorders. Presented at the 17th Annual BMT Tandem Meeting, Honolulu, HI, February 17-21, 2011. *Biol Blood Marrow Transplant*. 2011;17(S2):S173. Abstract 56.
2. Hurley CK, Klein JP, Spellman SR, et al. Scoring HLA mismatches by HistoCheck does not predict clinical outcome in HCT. Presented at the 17th Annual BMT Tandem Meeting, Honolulu, HI, February 17-21, 2011. *Biol Blood Marrow Transplant*. 2011;17(S2):S172. Abstract 55.

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3. Kawase T, Morishima Y, Malkki M, et al. Universal role for HLA-C and KIR 2DL ligand mismatch in severe acute GVHD after unrelated donor hematopoietic stem cell transplantation in Japanese and Caucasian transplant recipients: An analysis on behalf of the international Histocompatibility working group in HCT. Presented at the 17th Annual BMT Tandem Meeting, Honolulu, HI, February 17-21, 2011. Biol Blood Marrow Transplant. 2011;17(S2):S165. Abstract 37.
4. Venstrom JM, Pittari G, Chewning J, et al. Donor KIR2DS1 and KIR 3DS1 are associated with improved outcomes following unrelated allogeneic stem cell transplantation for acute myeloid leukemia. Presented at the 17th Annual BMT Tandem Meeting, Honolulu, HI, February 17-21, 2011. Biol Blood Marrow Transplant. 2011;17(S2):S156. Abstract 16.
5. Fleischhauer K, Spellman SR, Wang T, et al. Non-permissive HLA-DPB1 T-cell epitope disparities are associated with non-relapse mortality after unrelated stem cell transplantation and are not dependent on HLA-DPA1. Presented at the 37th Annual EBMT Meeting, Paris, France, April 3-6, 2011. Bone Marrow Transplant. 2011;46(S1):S74. Abstract O406.
6. Rocha V, Purtill D, Zhang M, et al. Impact of matching at non-inherited maternal antigens (NIMA) on outcomes after 5/6 or 4/6 HLA mismatched unrelated cord blood transplantation for malignant haematological diseases. A matched pair analysis on behalf of Eurocord, NetCord, NMDP, IBMTR. Presented at the 37th Annual EBMT Meeting, Paris, France, April 3-6, 2011. Bone Marrow Transplantation. 2011;46(S1):S2. Abstract O115.
7. Fleischhauer K, Wang T, Spellman SR, et al. No apparent contribution of HLA-DPA1 to the significantly increased risk for non-relapse mortality associated with non-permissive donor-recipient HLA-DPB1 T cell epitope disparities in unrelated stem cell transplants facilitated through the National Marrow Donor Program. Presented at the 2011 EFI Meeting, Prague, Czech Republic, May 3-6, 2011. Tissue Antigens 77(S5): 397. Abstract P4.
8. Dobbelsstein C, Haagensohn M, Ahn KW, et al. Birth order is not a major factor influencing transplant outcome in HLA-identical sibling SCT: an analysis on behalf of the CIBMTR. Presented at the 2011 European Hematology Association Meeting, London, June 9-12, 2011.
9. Brady C, Brown M, Eapen M, et al. The influence of HLA antigen/allele frequency on access to non-inherited maternal antigens (NIMA) for recipients of cord blood transplant. Best Poster Abstract. Presented at the 9th Annual Umbilical Cord Blood Symposium, San Francisco, June 23-25, 2011.

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10. Battiwalla M, Ellis K, Pavletic SZ, et al. HLA DR15 Antigen Status Does Not Impact Graft-Versus-Host Disease or Disease-Free Survival in HLA-Matched Sibling Transplantation for Hematologic Malignancies. Presented at the 53rd Annual ASH Meeting, San Diego, CA, December 9-13, 2011. Blood. 118(21): Abstract 3094.

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Attachment A – References

1. Spellman S, Hurley CK, Brady C, et al. Guidelines for the development and validation of new potency assays for the evaluation of umbilical cord blood. *Cytotherapy* 2011; 13(7): 848-855.
2. Kempenich JH, Dehn J, Michelle S. Inaccuracy of DRB1*16:08. *Tissue Antigens* 2012; 79(5): 449. Abstract P41.
3. Kempenich J, Dehn J, Flickinger G, et al. Rare allele typing project. *Human Immunol* 2010; 71(Suppl 1): Abstract 137-P.
4. Kempenich JH, Dehn J, Flickinger G, et al. Functional validation of rare HLA alleles. *Tissue Antigens* 2011; 77(5): 417. Abstract P54.
5. Dehn J, Maus M, Buck K, et al. A/B Only Typed Donors: Possible But Improbable 6/6 HLA Allele Matches. *Biol Blood Marrow Transplant* 2012; 18(2): S270-271. Abstract P180.
6. Kempenich JK, Howe K, Maus T, et al. AB only donors an untapped resource. *Human Immunol* 2012; 73(s1): Abstract 154-P.
7. Dehn J, Buck K, Yang SY, et al. 8/8 High-Resolution HLA Match Rate: The Impact of Race. *Biol Blood Marrow Transplant* 2011; 17(s2): S170-S171. Abstract O51.
8. Dehn J, Buck K, Setterholm M, Yang SY, Schmidt A, Confer D. Efficiency of strategic donor selection to identify an 8/8 high resolution match. *Tissue Antigens* 2011; 77(5): 408 (Abstract).
9. Xiao Y, Lazaro AM, Masaberg C, et al. Evaluating the potential impact of mismatches outside the antigen recognition site in unrelated hematopoietic stem cell transplantation: HLA-DRB1*1454 and DRB1*140101. *Tissue Antigens* 2009; 73(6): 595-8. .
10. Cooley S, Weisdorf DJ, Guethlein LA, et al. Donor selection for natural killer cell receptor genes leads to superior survival after unrelated transplantation for acute myelogenous leukemia. *Blood* 2010.
11. Cooley S, Trachtenberg E, Bergemann TL, et al. Donors with group B KIR haplotypes improve relapse-free survival after unrelated hematopoietic cell transplantation for acute myelogenous leukemia. *Blood* 2009; 113(3): 726-732.

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Attachment B – Published Manuscripts and Abstracts
Associated with this Grant

Manuscripts and Book Chapters

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